8 September 2006 | $10

# Science

AAAS

Volume 313, Issue 5792


## COVER

Part of the exposed western scarp at Erebus crater, within Meridiani Planum, Mars, showing tilted, stratified bedrock 1 to 2 m thick. These rocks contain textures indicative of sedimentary processes, as described on page 1403. The image was acquired by the Pancam instrument onboard the Mars Exploration Rover Opportunity on 2 March 2006; this false-color composite was generated from Pancam's 750-, 530-, and 430-nm filters.

***Photo: NASA/JPL/Cornell***

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## BOOKS *ET AL.*



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www.sciencexpress.org

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**The Consensus Coding Sequences of Human Breast and Colorectal Cancers**

*T. Sjöblom* et al.

Sequence analysis of >13,000 genes in breast and colorectal tumors shows that almost 200, a surprisingly large number, can be mutated, complicating any simple classification.

*>> News story p. 1370*

10.1126/science.1133427

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**Unraveling the Mystery of Indian Monsoon Failure During El Niño**

*K. Krishna Kumar, B. Rajagopalan, M. Hoerling, G. Bates, M. Cane*

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10.1126/science.1131152

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*M. Selmer* et al.

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10.1126/science.1131127

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10.1126/science.1132319

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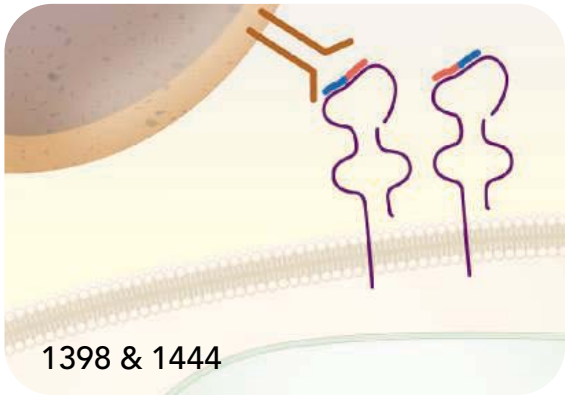
## BREVIA

PSYCHOLOGY





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## REPORTS

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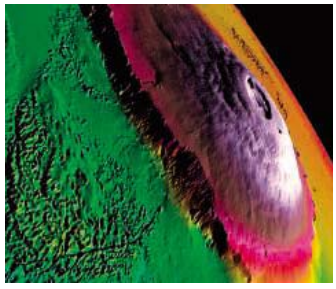

Tipping the scales.

## SCIENCE**NOW**

www.sciencenow.org DAILY NEWS COVERAGE

**Nerves Conquer Pain**
Blocking an enzyme in the spinal cord reduces pain and inflammation in arthritic rats.

**Earth's Poles May Have Wandered**
Large mass may have caused planet to "rebalance" itself 800 million years ago.

**Flashing Out a Star's Demise**
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**MISCINET: Policy Issues and Emotions**
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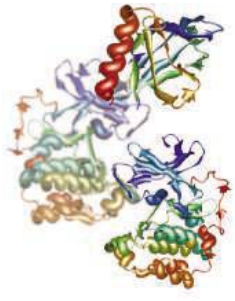


Structures of AGC kinases.

## SCIENCE'S **STKE**

www.stke.org SIGNAL TRANSDUCTION KNOWLEDGE ENVIRONMENT

**REVIEW: Systems Biology of AGC Kinases in Fungi**
*A. Sobko*
Is Sch9 the yeast homolog of protein kinase B?

**ST ON THE WEB: Cancer Genome Anatomy Project**
Explore the genes that contribute to cancer; in Bioinformatics Resources.

**ST ON THE WEB: DAVID—Database for Annotation, Visualization and Integrated Discovery**
Analyze microarray and proteomic data with these free online tools; in Bioinformatics Resources.

EDITED BY STELLA HURTLEY AND PHIL SZUROMI

## Where's Which Whisker?

Passing through several relay stations in the brain, sensory signals from the face are received in the somatosensory cortex of the brain in a spatial organization roughly reflecting that of the signal's origins. **Oury *et al.*** (p. 1408, published online 10 August) now show that in one of the relay stations in mice, the PrV nucleus, expression of *Hox* genes during development helps maintain the map and allows, for example, the discrimination of signals from the whiskers, upper jaw, and lower jaw.

## Water on Terrestrial Planets

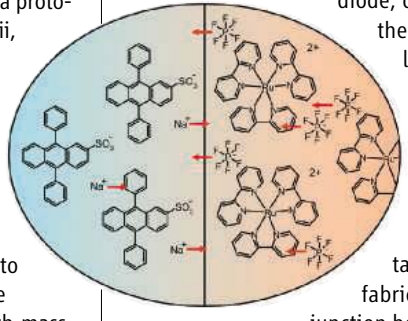
The Mars Exploration Rover Opportunity recently traveled 8 kilometers across Meridiani Planum, and an analysis by **Squyres *et al.*** (p. 1403; see the cover) of the features that it discovered has revealed information about ancient environmental conditions. These features include cross-laminations that formed in flowing liquid water, strata with hematite-rich concretions, weathered rock rinds, and networks of polygonal fractures likely caused by dehydration of sulfate salts. Chemical alteration of basalt can explain the composition of a 7-meter stratigraphic section. Observations from microscopic to orbital scales reinforce the conclusion that ancient Meridiani was characterized by abundant acidic groundwater, arid and oxidizing surface conditions, and occasional liquid flow on the surface. Beyond our solar system, some of the giant gas planets that have been observed have orbits that are much closer to their central stars compared to that of Jupiter in our own solar system. As gas giants should form from leftover gas in a protoplanetary disk more readily at large radii, they must gradually spiral inward, but this process would disrupt any other planets in that system. **Raymond *et al.*** (p. 1413) have simulated the behavior and formation of Earthlike planets in systems where a gas giant migrates inward and show that terrestrial planets can still form both interior and exterior to the migrating jovian planet. Outside the giant planet's orbit, very water-rich earth-mass planets could form within the habitable zone.

## High and Dry

The vegetation of Eastern Africa shifted progressively from forest to grassland between 8 and 2 million years ago, and this change has been ascribed to the influence of decreasing concentrations of atmospheric $CO_2$ (which favors grasses over trees), recurring periods of aridity caused by changing sea surface temperatures, and the beginning of glacial cycles. **Sepulchre *et al.*** (p. 1419) suggest that another contributing factor could have been increasing aridity caused by tectonic uplift along the East African Rift System, which would have led to a dramatic reorganization of atmospheric circulation and a strong drying trend. They examine the climatological and biological effects of uplift through numerical modeling, and conclude that it must have been a dominant factor in determining late Neogene African climate.

## Ionic Electroluminescence

In a classic *pn*-junction between *n*-type and *p*-type semiconductors, the transfer of an electron through the junction can cause emission of light, as in a light emitting diode, or conversely, the absorption of light can lead to an electric current, as in a solar cell. **Bernards *et al.*** (p. 1416) used soft-contact lamination to fabricate an ionic junction between two organic semiconductors with mobile anions and cations. Similar to the classic *pn*-junction in which electrons are the mobile species, ionic charges can be successfully used to control the direction of electronic current flow in these semiconductor devices, which show electroluminescence under forward bias and produce a photovoltage upon illumination with visible light.



## Solid-State Entanglement

Entanglement between qubits is a necessary requirement for any proposed quantum computer architectures, and solid-state implementations, particularly superconducting qubits, have the added advantage of being compatible with existing fabrication techniques. To date, the behavior and manipulation of single superconductor-based qubits have shown promising results. **Steffen *et al.*** (p. 1423; see the Perspective by **Siddiqi and Clarke**) use state tomography to demonstrate that entanglement between two superconducting phase qubits is possible. These new results put solid-state qubits on the roadmap as a basis for a scalable quantum computer.

## Volcanic Cracks in the Ocean Floor

Volcanism on Earth occurs at plate boundaries (such as mid-ocean ridges and island arcs) and within plates above mantle plume hot spots. **Hirano *et al.*** (p. 1426, published online 27 July; see the Perspective by **McNutt**) report finding another type of volcano that is far from any of these primary sources. In submersible dives in the western Pacific Ocean, far from the plate edge, they saw the tops of small volcanoes that were partly buried in sediment and surrounded by pillow lavas and exploded shards. Geochemical analysis suggests the resulting basalts are young and formed at depths greater than 100 kilometers in the asthenosphere, which would imply that this layer contains a few percent melt. The authors

CREDITS (TOP TO BOTTOM): XAVIER LAMPE; BERNARDS *ET AL.*

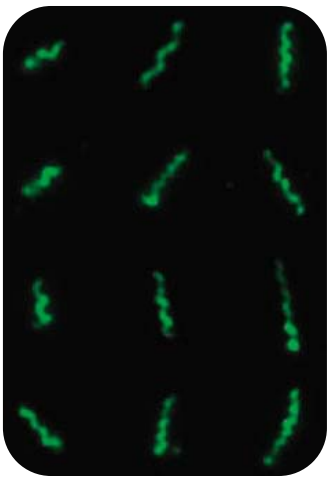
argue that these "petit spot" volcanoes have grown along cracks where the asthenosphere has flexed and squeezed out its melt.

## Of Mice and Men and Immunity

The immunity-related p47 guanosine triphosphatases are a class of innate immunity effectors found in murine cells where they play a role in defense against intracellular pathogens. However, the role of similar proteins in humans has been less clear. Now **Singh *et al.*** (p. 1438, published online 3 August) demonstrate that in mouse cells one of these receptors acts via autophagy, inducing large autolysosomal organelles to destroy intracellular *Mycobacterium tuberculosis* bacilli. Furthermore, the sole human counterpart, IRGM, also works via autophagy to control intracellular mycobacteria.

## The Humanization of Yeast

The ability to produce proteins modified with humanlike carbohydrates is important in therapeutics and structural studies. **Hamilton *et al.*** (p. 1441) describe the genetic engineering of the secretory pathway of the yeast *Pichia pastoris* to produce structurally homogeneous complex, terminally sialylated human-type N-glycans on therapeutically efficacious erythropoietin. The engineered cell lines contain a total of four gene knockouts and 14 heterologous genes, the majority of which had not been identified in nature and had to be discovered through an extensive screening effort.



CREDIT: YOSHIDA *ET AL.*

## Dissecting Chloroplast Division Machinery

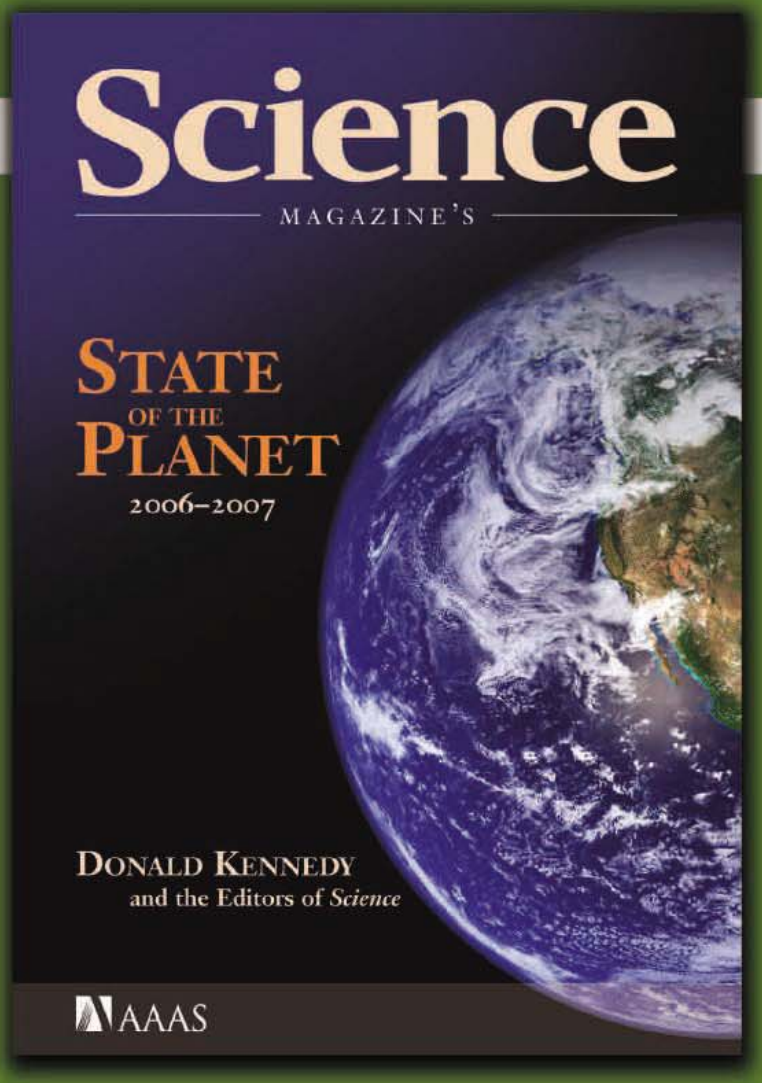
Chloroplasts arose from an endosymbiotic cyanobacterial ancestor and have their own genomes that have been maintained by division. **Yoshida *et al.*** (p. 1435) isolated intact circular chloroplast division machineries containing dynamin and FtsZ from the red alga *Cyanidioschyzon merolae*. Rings isolated at the early phase of division formed supertwisted (or spiral) structures that could be reversibly stretched to four times their original length with optical tweezers. As the contraction of the rings progressed, small compact circles were produced, and the dynamin pinched off the narrow bridge between daughter chloroplasts. Thus, dynamin may function both as a mediator of filament sliding and as a pinchase during chloroplast division.

## Making Even More Diversity

Recently, a role for the proteasome was discovered in splicing together noncontiguous peptides into effective antigens. **Warren *et al.*** (p. 1444; see the Perspective by **Shastri**) identified an antigenic peptide that corresponds to a minor histocompatibility antigen that is expressed on leukemic cells. The antigen was also created in the proteasome by splicing of two noncontiguous fragments of the parental protein, but the two fragments were spliced in the reverse order to that in which they occur in the parent protein. Splicing of these reordered peptide fragments occurred by transpeptidation involving an acyl-enzyme intermediate. This mode of production of antigenic peptides expands the diversity of antigenic peptides presented on class I molecules and is potentially relevant for T cell recognition of tumors and pathogens.

## Clean Bodies, Clean Minds

Cleanliness is regarded as a desirable state, not only in the physical sense of personal hygiene but also in the moral sense of feeling virtuous. **Zhong and Liljenquist** (p. 1451) describe a sequence of studies that make the connection between physically washing one's hands and feelings of virtue. Ethically compromised individuals experienced an increased desire to cleanse themselves, but physical cleansing alleviated the psychological consequences of unethical behavior, both assuaging moral emotions and reducing moral-compensatory behavior.




*Science* Magazine's

## State of the Planet 2006-2007

Donald Kennedy, Editor-in-Chief, and the Editors of *Science*

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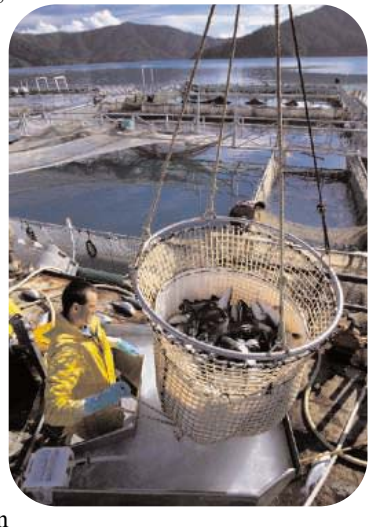
Rosamond Naylor is the Julie Wrigley Senior Fellow at the Freeman-Spogli Institute for International Studies and the Woods Institute of the Environment at Stanford University, and the director of Stanford's program on Food Security and the Environment.

# Offshore Aquaculture Legislation

FISH FARMING IS FLOURISHING ALONG COASTLINES IN MANY COUNTRIES. BUT THE United States is turning instead to the open ocean for aquaculture expansion. The National Oceanic and Atmospheric Administration (NOAA), a unit within the U.S. Department of Commerce, justifies this move on several grounds: America's seafood appetite continues to grow, ocean waters are overfished, and marine fish farming near the shore is limited by state regulations. As a result, the United States faces a large and growing seafood deficit, now around $8 billion annually. With technology such as submersible cages with robotic surveillance becoming available for open-ocean farming, why not move aquaculture into the high seas? After all, the United States has the largest exclusive economic zone (EEZ) in the world, amounting to roughly 1.5 times the landmass of the lower 48 states. Facilitating aquaculture development in federal waters of the EEZ (3 to 200 miles offshore) could result in substantial commercial benefits. But at what cost to sustainable fisheries, wild fish populations, and marine ecosystems remain sticky questions for legislation.



On 8 June 2005, Commerce Committee Co-Chairmen Senators Ted Stevens (R-AK) and Daniel Inouye (D-HI) introduced the National Offshore Aquaculture Act of 2005 (S. 1195). This bill, crafted by NOAA, establishes a permitting process for offshore aquaculture development within the federal waters of the EEZ and encourages private investment in aquaculture operations, demonstrations, and research. It gives the Secretary of Commerce the authority and broad discretion to promote offshore aquaculture—in consultation with other relevant federal agencies, but without firm environmental requirements apart from existing laws. Just how much NOAA should be promoting versus overseeing aquaculture development is debatable, particularly because many of the needed environmental safeguards are missing. Without a clear legal standard for environmental and resource protection within the bill, marine fisheries and ecosystems are vulnerable to further decline.

Ample evidence from near-shore systems indicates major environmental risks from fish farming: The escape of farmed fish from ocean cages can have detrimental effects on wild fish populations through competition and interbreeding, parasites and diseases can spread from farmed to wild fish, there is damaging nutrient and chemical effluent discharge from farms, and the use of wild pelagic fish for feed can deplete the low end of the marine food web in certain locations. Species targeted for offshore systems, such as halibut and cod, are also caught in the wild, so commercial fishing interests worry about the economic as well as ecological consequences. Most existing open-ocean systems are experimental. They experience predator attacks, escapes, and high use of wild fish for feed, and the full ecological impact of commercial-scale offshore aquaculture remains unknown.

Since the introduction of S. 1195, environmental and fishing groups have worked hard to stop the legislation. The bill was roundly criticized before a Senate committee in June 2006 and has yet to reach the House. In the likely event that S. 1195 resurfaces in the next legislative session, stakeholders and the public should be attentive to three points. First, states have an important role to play. For example, California's recent Sustainable Oceans Act (SB 201) sets high environmental standards for marine finfish production in state waters and could help shape national legislation. An amendment to S. 1195 also permits states to opt out of aquaculture development in federal waters off their shores. Second, industry leaders whose business strategy strongly incorporates environmental and social stewardship should contribute to the bill's revision. Positive participation by the industry would help move the legislative process forward. Finally, the revised legislation must permit firms operating in U.S. federal waters to be internationally competitive. This will only happen if the bill is crafted in an international context, with sound environmental standards adopted in all countries with marine aquaculture, whether near shore or offshore. Commerce is eyeing the global picture. So too should the global environmental community.

**– Rosamond Naylor**

10.1126/science.1134023

The hawksbill turtle.

ECOLOGY/EVOLUTION

## Sex on the Beach

For many reptiles, the temperature at which their eggs are incubated determines the sex of the hatchling. In a world affected by global climate change and localized anthropogenic pressures, temperature-dependent sex determination can have all-or-none consequences for sex ratios and hence population viability. Kamel and Mrosovsky document a graphic example of this peril, in the case of the hawksbill turtle in the Caribbean. Like other marine turtles, hawksbills lay their eggs above the high tide mark on beaches. Where the beach is shaded by its natural forest cover, cooler incubation temperatures lead to a more male-biased sex ratio. However, such male-producing sites are increasingly scarce as more of the coastlines of Caribbean islands are deforested and developed for tourism, and there is evidence that the hawksbill population is becoming more female-biased. — AMS

*Ecol. Appl.* **16**, 923 (2006).

CHEMISTRY

## A Convenient Couple

Biaryls are a common structural motif in pharmaceutically important compounds and have traditionally been prepared using strategies that couple a halogenated substrate to a second compound pre-adorned with a reactive group such as a boronic ester or alkyl stannane. Recent research has focused on improving the efficiency of these syntheses by linking aryl halides directly to the aromatic C-H bond of a partner ring. Yanagisawa *et al.* extend this trend with a rhodium catalyst that couples iodobenzene and its derivatives efficiently to heterocyclic aromatics, including substituted thiophenes, furans, and pyrroles. At 3 mole % loading, the catalyst induces regioselective bond formation at the carbon adjacent to an oxygen or sulfur atom, though somewhat surprisingly selects for the 3 position in N-substituted 1-phenylpyrrole. Pi-accepting bulky phosphite ligands played a crucial role in achieving catalytic efficiency and also conferred air stability on the Rh complex. The catalyst proved capable of coupling aryl halides to methoxy-substituted benzenes as well, albeit with diminished regioselectivities relative to those obtained with the heterocyclic substrates. — JSY

*J. Am. Chem. Soc.* **128**, 10.1021/ja064500p (2006).

ASTROPHYSICS

## Polarized Snaps

Buried in the patterns of the cosmic microwave background radiation that bathes the sky are clues to the structure of the universe. Ripples in temperature have been mapped in fine detail for several years, but further insight requires the mapping of polarized signatures that place extra constraints on early-universe physics. One pioneering experiment that has measured temperature anisotropies is BOOMERanG—Balloon Observations Of Millimetric Extragalactic Radiation and Geophysics—a balloon-borne array of bolometer detectors floated from Antarctica. In a 200-hour flight in January 2003, BOOMERanG succeeded in mapping detailed structures in polarized light at 145 GHz over a few percent of the full sky. In a series of papers, MacTavish *et al.*, Montroy *et al.*, Jones *et al.*, and Piacentini *et al.* report the latest power spectra determinations of temperature, polarization, and temperature polarization cross-correlations. These results are consistent with recent measurements on degree scales by the Wilkinson Microwave Anisotropy Probe (WMAP) satellite but also extend to much higher resolution and offer finer sampling than has been achieved to date by other low-frequency experiments. The BOOMERanG data are consistent with the consensus cosmological model, a universe dominated by dark energy and cold dark matter. Some models of early structure formation are ruled out, notably defects, and adiabatic seed fluctuations are favored. — JB

BOOMERanG launch.

*Astrophys. J.* **647**, 799; 813; 823; 833 (2006).

BEHAVIOR

## Learning to Lift or Slide

Evidence for the cultural transmission of behaviors in nonhuman primates comes primarily from long-term observational histories of wild populations. To counter the criticism that theories derived from these data sets are inference-based, Horner *et al.* describe an experimental study demonstrating that a naïve chimpanzee can figure out how to forage for food by watching a skilled practitioner and can then serve as a tutor for a third individual, creating a chain of learning. They designed a "Doorian fruit" box from which food could be retrieved by either lifting or sliding a door. When untutored chimpanzees (or 3-year-old children in a parallel series of trials) were presented with the apparatus, about half discovered how to open the door, some by lifting it and others by sliding it (which required equally effortful actions). On the other hand, when socially compatible chimpanzees were allowed to play the roles of teacher and student in strictly binary interactions, the initial mode of foraging (lift versus slide) was faithfully passed along a chain of individuals (six and five, respectively); a similarly exclusive transmission of the original foraging technique (for acquiring a toy) was found in chains of eight children. — GJC

*Proc. Natl. Acad. Sci. U.S.A.* **103**, 13878 (2006).

CREDITS (TOP TO BOTTOM): N. MROSOVSKY; BOOMERANG COLLABORATIONS

CLIMATE SCIENCE

## The Times Temps Were a'Changin'

The occurrence of several large and abrupt climate changes dated to the last deglaciation, first clearly evidenced in Greenland ice cores, has also been confirmed by a variety of other proxies in lower-latitude Northern and Southern Hemispheric marine and terrestrial records. Despite much knowledge of the environmental changes that accompanied these events, an understanding of their causal mechanisms is hampered by the difficulty of determining the absolute ages of the different records. In order to better determine the phase relationships of these events at different locations, Genty *et al.* analyzed stalagmite records of $\delta^{13}C$ isotopic distributions from several Northern Hemispheric locations, in France and Tunisia, and compared them with corresponding records from speleothems in China, New Zealand, and South Africa. The advantage of this approach is that stalagmites can be precisely dated, thereby establishing an accurate common chronology. The data suggest that the Bølling-Allerød warm interval began synchronously in France, Tunisia, and China; that the Younger Dryas cold period also began concurrently at all of these sites; and that although the onset times were the same at widely separated sites in both hemispheres, the duration and intensity of transitions differed among sites. The authors also suggest a simple explanation for these changes, involving the gradual increase of insulation at high northern latitudes, due to orbital changes, and the resulting northward movement of the limits of sea ice there. — HJS

**Dated stalagmite.**

CREDIT: GENTY *ET AL.*, *QUAT. SCI. REV.* **25**, 2118 (2006)

*Quat. Sci. Rev.* **25**, 2118 (2006).

CHEMISTRY

## A Different Sort of CP

To organometallic chemists, a "Cp" notation in molecular formulas is well understood to signify the widely used cyclopentadienyl ligand $C_5H_5$. The absence of confusion engendered by this abbreviation highlights the elusiveness of the cyaphide ligand CP: an analog of cyanide in which phosphorus replaces nitrogen. Cordaro *et al.* have succeeded in coaxing a precursor toward this long-sought diatomic and report isolation of a stable ruthenium complex coordinated to cyaphide through the carbon. Their synthetic route proceeds from a triphenylsilyl ($Ph_3Si$)–coordinated $CH_2PCl_2$ fragment to the $Ph_3Si$-C≡P phosphaalkyne through dehydrohalogenation. This molecule coordinates to a cationic Ru center to yield a stable complex that was characterized by x-ray crystallography. Addition of fluoride to a solution of this compound surprisingly led to attack at P rather than at the traditionally fluorophilic Si center. However, phenoxide proved a more cooperative nucleophile, liberating CP from the silyl cap. The resulting complex was characterized crystallographically and by nuclear magnetic resonance spectroscopy in solution; the vibrational spectrum revealed a C≡P stretching band at 1229 $cm^{-1}$. — JSY

*Angew. Chem. Int. Ed.* **45**, 10.1002/anie.200602499 (2006).

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www.stke.org

## << A Flexible Fate?

Specific factors in the local microenvironment govern the differentiation of bone marrow–derived mesenchymal stem cells (MSCs) into disparate cell types such as neurons, myoblasts, and osteoblasts, yet remain incompletely understood. Noting that brain tissue is much softer than muscle, which in turn is softer than collagenous bone, Engler *et al*. cultured naïve human MSCs on collagen-coated polyacrylamide gels in which elasticity was varied via the extent of bis-acrylamide crosslinking in order to investigate the role of matrix elasticity in lineage specification. The morphology, transcriptional profile, and expression of marker proteins of MSCs grown for a week on soft gels (mimicking brain tissue) resembled those of cultured neurons; MSCs grown on gels that mimicked the elasticity of striated muscle resembled myoblasts; and MSCs grown on gels that mimicked young uncalcified bone resembled osteoblasts. During the first week in culture, exposure to soluble factors known to promote myogenic or osteoblastic differentiation influenced lineage, leading to a mixed MSC phenotype. After 3 weeks in culture, however, MSCs remained committed to the matrix-derived lineage. Pharmacological analysis indicated that nonmuscle myosin II was required for lineage specification in response to matrix elasticity but not in response to soluble factors. Thus, the data suggest that matrix elasticity plays an important role in specifying MSC lineages. — EMA

*Cell* **126**, 677 (2006).

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See pages 102 and 103 of the 6 January 2006 issue or access www.sciencemag.org/feature/contribinfo/home.shtml

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IMAGES

## GO WITH THE FLOW

It isn't a fancy Rorschach blot or a computed tomography scan of the intestines. Instead, the image above depicts the chaotic mixing caused by stirring a vat of glycerin and fluorescent dye. It's one example of liquid artistry on display at this gallery* from the journal *Physics of Fluids*. Showcased here are winning entries from the American Physical Society's annual exhibition of videos and photos. You can admire shots from as far back as 1985, although you'll need a journal subscription to see the newest entries. This fluid dynamics collection† from applied mathematician John Bush of the Massachusetts Institute of Technology uses a strobe lamp and other tricks to reveal unexpected and striking patterns, such as the trail of turbulence created by a water strider. >>

* pof.aip.org/pof/gallery/index.jsp
† www-math.mit.edu/~bush/gallery.html

EXHIBIT

## Milky Way Portraitist >>

Staying up late paid off for American astronomer Edward Emerson Barnard (1857–1923). Dubbed "the man who never slept," the telescope virtuoso took gorgeous photos of our galaxy, such as the nebula of Rho Ophiuchi (right), and discovered a slew of heavenly objects, including Jupiter's fifth moon Amalthea. At this exhibit from the Georgia Institute of Technology in Atlanta, you can peruse Barnard's magnum opus, the posthumously published *Atlas of Selected Regions of the Milky Way*. Although he left school at age 9, the self-taught observer rose to be a professor at the University of Chicago and sat at the controls of the world's largest telescopes. Astronomers still value the atlas for its wide-angle views and because it revealed murky areas in space that eventually led to the discovery of dark matter. >>
www.library.gatech.edu/about_us/digital/barnard/index.html

WEB LOGS

## Small News

Microbe fans can get an eyeful of viruses or an earful of bacteria at the new educational Web log Microbiology Bytes from Alan Cann of the University of Leicester in the U.K. Along with written commentary, Cann offers excursions into the microbial world in the form of enhanced podcasts, which feature video and graphics as well as audio narration. Podcast topics include determining how many bacterial species dwell in the soil and recent studies on the use of RNA interference to block cold sores. >>
microbiologybytes.wordpress.com

DATABASE

## Framingham Gene Hunt

The race to find the genes behind common ailments is heating up as many research groups scan patients' entire genomes for markers linked to disease. When it opens later this month, the Genomic Medicine Database (GMED) from Boston University (BU) will showcase such results from 1320 participants in the famed Framingham Heart Study, which has followed the health of a small Massachusetts town for 50 years. You can peruse the chromosomes for possible associations between about 10 traits—such as hypertension and high cholesterol levels—and 100,000 genetic markers, known as SNPs. Click to zoom in on the genes near a SNP. The BU team is posting data before publication so that other researchers can quickly seek to replicate the findings, says GMED co-curator Marc Lenburg. "Our hope is that others will follow our lead" and share unpublished data, he says. >>
gmed.bu.edu

RESOURCES

## Flu on the Wing

This new avian influenza monitoring site houses no data on the highly pathogenic H5N1 virus in U.S. wild birds—but that's a good thing. As the U.S. Geological Survey clearinghouse records, none of the more than 11,800 birds sampled in 28 states so far this year carried the virulent strain, which experts fear could morph into a virus that triggers a pandemic. If the deadly virus does infect wild birds here, as it has done in Asia and Europe, visitors will be able to follow the results state by state. >>
wildlifedisease.nbii.gov/ai

**Send site suggestions to >>** netwatch@aaas.org
**Archive:** www.sciencemag.org/netwatch

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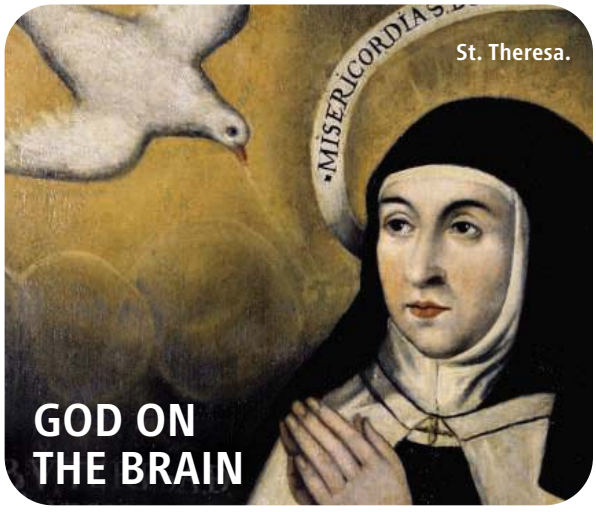



Nuns replaying past mystical experiences have made the latest contribution to the burgeoning field of "spiritual neuroscience."

Psychologist Mario Beauregard of the University of Montreal in Canada and his student Vincent Paquette recruited 15 Carmelite nuns, all of whom had had at least one intense mystical experience. The two researchers looked at the nuns' brains using functional magnetic resonance imaging while the sisters tried to re-evoke such experiences. As a control, the nuns' brains were also imaged while they tried to relive "the most intense state of union with another human" they had ever felt.

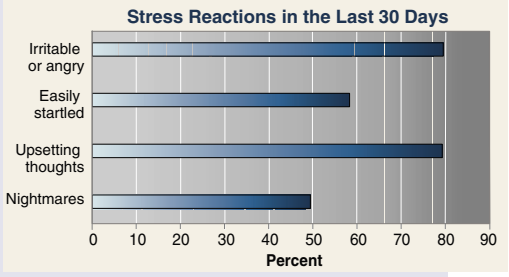
Beauregard says that some researchers have theorized that religious experiences involve epilepsy-like seizures in temporal lobes. But the mystical condition activated dozens of brain areas involved in perception, emotion, and cognition, he and Paquette reported last week in *Neuroscience Letters*. The pair also conclude that although there is much overlap with the feelings of peace and love from the control condition, the mystical condition has its own signature, with "relatively different regional patterns of brain activation."

Physician Andrew Newberg, head of the newly established Center for Spirituality and the Mind at the University of Pennsylvania, says the study indicates that a mystical state activates a larger brain area than would ordinarily be involved in focusing on a specific problem or memory, so such states are "extremely complex."

# Katrina's Mental Fallout

The incidence of serious mental illnesses among Hurricane Katrina survivors doubled within 5 to 8 months after the storm, according to a telephone survey by epidemiologists at Harvard Medical School in Boston. But the study found a surprising absence of suicidal tendencies among the survivors.

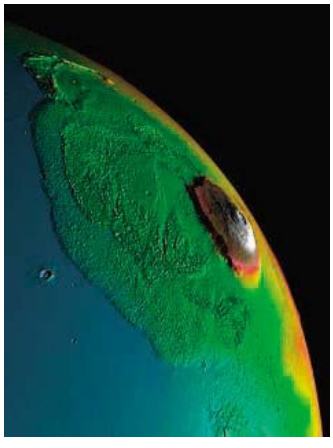
The researchers interviewed 1043 survivors between 19 January and 31 March about their post-Katrina experiences and documented that 30% had mental-health problems, half of them serious—a doubling of the rate seen in a face-to-face survey conducted between 2001 and 2003.




Problems such as anxiety and nightmares among New Orleans residents (see chart) were more frequent than among hurricane survivors elsewhere. Project director Ronald Kessler said the findings show that many "have a level of [mental] disorder that is going to interfere with the rebuilding of their lives." Most (84.6%) had lost their housing and income, and 36.3% had experienced severe physical hardship, including hunger. Of the 40.6% who experienced five or more stressors, such as property loss, physical hardship, or losing a loved one, close to half were in the bottom 25% of income level.

But despite the problems, suicidal tendencies had decreased since the storm: Only 0.4% reported such thoughts compared to 3.6% in the earlier survey. The researchers attribute this to a sense of personal growth following the disaster. For example, 88.5% reported developing a deeper sense of meaning or purpose in life, and 83.4% were confident in their ability to rebuild their lives.

Scientists will continue to track the group over the next few years.



**Polar wander on Mars was caused by large volcanoes.**

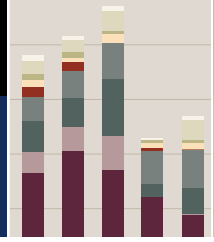
## << WANDERING POLES

New findings support an old but controversial theory that Earth's poles have on occasion made gigantic shifts in their placement. Such major relocations, known as "true polar wander," are believed to result from changes in weight distribution on a planet's surface, such as those caused by a huge volcanic eruption. This would cause the planet to realign itself in relation to its spin axis, moving the poles.

Evidence that Earth's poles shifted dramatically about 800 million years ago has been found in magnetic rocks in Australia and China. Now, a team led by geologists Adam Maloof of Princeton University and Galen Halverson of Paul Sabatier University in Toulouse, France, have added data from Norwegian rocks. As magnetic mineral grains were deposited or excreted by microbes in the rocks, they aligned themselves with Earth's magnetic field, becoming frozen compasses pointing to an ancient north pole. Maloof and Halverson estimated from a stack of deposits laid down over the course of 20 million years that during that time, the north pole shifted more than 50 degrees—about the distance between Alaska and the equator.

The paper, published in the September-October issue of the *Geological Society of America Bulletin*, is an "important one," says geologist Rob Van der Voo of the University of Michigan, Ann Arbor, and it will help scientists determine how the continents fit together in the ancient supercontinent Rodinia.

A boost for Germany 1373



No end to earmarks 1374

CANCER

# First Pass at Cancer Genome Reveals Complex Landscape

Scientists have long known that the sparks that kindle cancer are mutations in a cell's genes. But most cancer-causing mutations have been discovered by looking in obvious places, such as in the genes that control cell division. Now it seems these efforts have barely glimpsed the big picture.

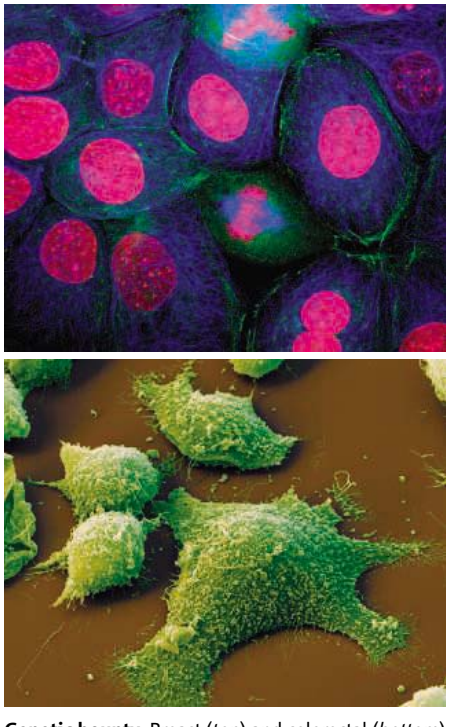
As reported online this week in *Science* (www.sciencemag.org/cgi/content/abstract/1133427), researchers have shined a searchlight across the genomes of breast and colorectal cancer cells, looking for mutations in more than half of all known human genes. And what they've uncovered is a much larger and richer set of cancer genes than expected.

The findings, hailed as a tour de force by other cancer scientists, should speed the race for new drugs, diagnostics, and a better understanding of tumor development. "It will take a long time to unravel all of this, but this is what cancer *is*," says Bert Vogelstein of Johns Hopkins Kimmel Cancer Center in Baltimore, Maryland, a co-leader of the sequencing effort.

The results also appear to bolster The Cancer Genome Atlas, an ambitious $1.5 billion federal project to systematically search for genes mutated in dozens of cancer types (*Science*, 29 July 2005, p. 693). "I see this as a big shot in the arm for the argument that this strategy is going to work," says Francis Collins, director of the National Human Genome Research Institute in Bethesda, Maryland, which together with the National Cancer Institute (NHGRI) will soon announce details of a $100 million, 3-year pilot effort for the atlas. Adds Eric Lander, director of the Broad Institute in Cambridge, Massachusetts, who first proposed sequencing the cancer genome, "This is a beautiful demonstration that if you turn over every rock, there is a lot more to be found."

Yet even supporters of the atlas say this first, quick pass at describing all cancer mutations reveals daunting complexity. And not everyone has been convinced of the larger project's value. Geneticist Stephen Elledge of Harvard Medical School in Boston, while predicting that the new study will become a "classic paper," says that a costly sequencing project will give short shrift to functional genomics studies and take money away from investigators working on equally important cancer efforts. "I still believe we need a more balanced approach," says Elledge, who first expressed those concerns last year (*Science*, 21 October 2005, p. 439).

To conduct this mini–cancer-genome project, a 29-person team, headed by Vogelstein and Hopkins colleagues Kenneth Kinzler and Victor Velculescu, began with a database of 13,023 genes that are considered the best-studied and annotated of the 21,000 known genes in the human genome. Led by postdoc Tobias Sjöblom, the team resequenced the protein-coding regions of the genes in 11 breast cancer samples and 11 colon cancer samples, yielding 800,000-plus possible mutations. The team then winnowed out more than 99% of the mutations by removing errors, normal variants, and changes that didn't alter a protein.



**Genetic bounty.** Breast (*top*) and colorectal (*bottom*) cancer cells contain many mutated genes.

They ultimately found that the average breast or colon tumor has 93 mutated genes, and at least 11 are thought to be cancer-promoting. This yielded a total of 189 "candidate" cancer genes. Although some are familiar—the tumor-suppressor gene *p53*, for example—most had never been found mutated in cancer before. And the abundance of certain types of genes, such as those involved in cell adhesion and transcription, suggested that these processes play a huge role in cancer. The results, says Ronald DePinho of the Dana-Farber Cancer Institute in Boston, are a "treasure trove."

Verifying that each candidate gene is important to cancer won't be simple. Not only did the cancer genes differ between colon and breast cancers, but each tumor had a different pattern of mutations. The number of genes suggests that there may be more steps to cancer than thought. "It's a much more complex picture than we had anticipated," Vogelstein says.

At least two other pilot cancer-genome projects—one funded by NHGRI and one led by Michael Stratton and P. Andrew Futreal of the Sanger Institute in Hinxton, U.K.—are yielding similar results. The Sanger effort is looking at 500 genes in a larger number of tumor samples and cancer types and, according to an e-mail from Stratton and Futreal, has also found a "tremendous diversity of mutation number and pattern between cancers."

DePinho says the mutation differences from tumor to tumor could help explain why 90% of drugs fail in patients. Elledge, for his part, says the relatively small number of new genes common to the tumors reinforces his concerns about The Cancer Genome Atlas. He suggests that some of the government's money would be better spent on more direct studies, such as screens for lethal genes in cancer cells. The cost of the Hopkins study alone—Vogelstein says it took about $5 million, mostly from private funding sources—could fund five National Institutes of Health (NIH) grants on such topics, Elledge notes.

Despite such doubts, the atlas project gets under way next week. NIH will announce the three cancers to be studied in the pilot phase and a set of repositories that will supply tissue samples for sequencing. Centers that will characterize the genes will be announced in early October. The project is on an "extremely aggressive timeline," says DePinho, who co-chairs its advisory committee. **–JOCELYN KAISER**

EUROPEAN SCIENCE

# Basic Science Agency Gets a Tag-Team Leadership

**BERLIN—**In a surprise decision, Europe has selected two leaders as successive heads of its new basic science agency, the European Research Council (ERC). The governing council announced last week that it has chosen biochemist Ernst-Ludwig Winnacker, current president of the German funding agency DFG, to be secretary general of ERC, which will make its first awards next year. But in July 2009, halfway through the 5-year term, Winnacker will be succeeded by Spanish economist Andreu Mas-Colell, who will serve through 2011.

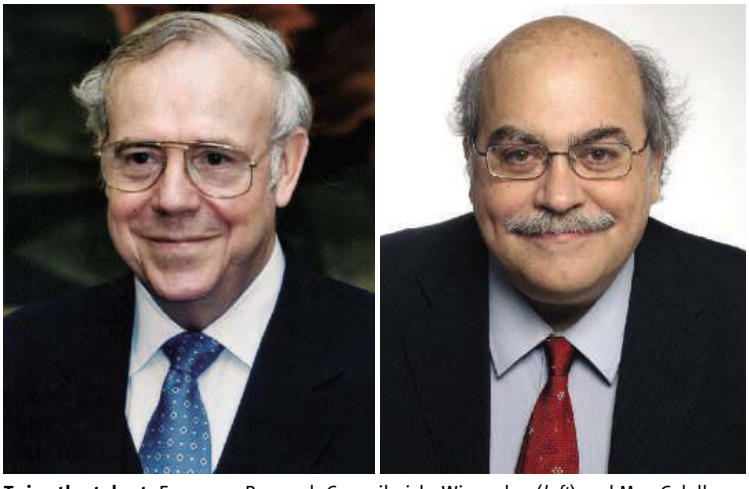
Members of ERC's board said they created the unusual arrangement to recruit executives with different skills, not because either candidate requested a short appointment. "We couldn't pass up these two exceptional people who are very complementary," says scientific council chair Fotis Kafatos, a molecular geneticist at Imperial College London. "Either one would have been great; having both will be even greater."

ERC is designed to be a sort of National Science Foundation (NSF) for all of Europe, and its $9.6 billion budget over 7 years is expected to fund cutting-edge research. But as the European Union–backed initiative gets off the ground, it faces a legacy of red tape in European science funding. Researchers have high hopes that it will prove much more user-friendly than the previous R&D efforts, called "Framework" programs, roundly criticized for the mountains of paperwork they generate. Kafatos says one early triumph is ERC's ability to make awards as research grants instead of the complicated contracts that other E.U. funding schemes require.

The ERC Scientific Council, made up of 22 leading scientists from across Europe, sets ERC's rules and scientific guidelines. The secretary general will be ERC's chief executive, serving as a liaison between the Scientific Council and the European Commission, which will handle day-to-day operations.

Both Winnacker and Mas-Colell say they were surprised to learn that they would serve truncated terms, which they were informed of at the same time they received the job offer, but both said they were honored to be chosen. Science Council vice-chair Helga Nowotny of the Vienna Centre for Urban Knowledge Management says the arrangement is intended to take advantage of the strengths of both men. In its start-up phase, she says, ERC needs someone with extensive experience overseeing a large granting organization. That's what Winnacker has done at DFG. But it will also need someone to stump for increased funding and to deal with politicians who may be unhappy with grants awarded on the basis of excellence without regard to geographic distribution. Mas-Colell's credentials as an economist and former state research minister will help him make a persuasive case, Nowotny says: "I think we will make good use of both of them, and we need both of them."

Winnacker, 65, had already announced plans to step down as DFG president at the end of 2006. This is "a solid appointment of someone who knows how to manage science at the highest level," says Frank Gannon of the European Molecular Biology Organization in Heidelberg, Germany. Winnacker's experience at the semiautonomous DFG makes him well positioned to fight for ERC's independence if challenged by the E.U. Parliament or member country politicians, Gannon says: "He will not be pushed around."



**Twice the talent.** European Research Council picks Winnacker (*left*) and Mas-Colell.

Mas-Colell, 62, is a professor at the University Pompeu Fabra in Barcelona, president of the European Economic Association, and was commissioner for universities and research for Catalonia from 1999 to 2003. He is credited with fostering science investment in the region, which led to the development of several new institutes in Barcelona (*Science*, 2 June, p. 1295). Mas-Colell spent 26 years at the University of California, Berkeley, and Harvard University before returning to Spain in 1995. Last year, he told a meeting of economists to judge ERC's success on how closely it emulated the U.S. NSF. He says now that he was thinking especially of NSF's widely praised peer-review system.

The scientific council's first call for applications will target young scientists, with 5-year awards of €100,000 to €400,000 per year. It hopes to award 200 such grants annually. A second program will target "advanced investigators" in a program intended to overcome both the limited size of awards given by national councils and the E.U.'s requirement that large projects be divided among many countries.

"One of the weaknesses of the European system is that most of the national [funding] councils are too small to fund their excellent scientists adequately," Winnacker says. But until now, large collaborative projects typically have required investigators from multiple countries. Winnacker says ERC's freedom from such geographical constraints will be "a big step forward. … No one would require someone from Massachusetts to collaborate with someone from South Dakota." **–GRETCHEN VOGEL**

CREDITS (LEFT TO RIGHT): DFG; CENTRUM FOTOŠKODA

FDA

# Proposed Guidelines for Emergency Research Aim to Quell Confusion

Doing research in the emergency room would be difficult even if the rules were clear, but many clinicians say they aren't. Last week, the U.S. Food and Drug Administration (FDA) suggested revisions to its regulation over an ethically fraught but critical area: studies conducted in emergency situations, when subjects may be unconscious and unable to give consent. The current 10-year-old FDA rule permits emergency research under narrow circumstances—in life-threatening medical conditions in which available treatments are unsatisfactory.

**Under review.** Research in emergency situations, which raises tough ethical questions, is receiving FDA scrutiny.

Hoping to clarify the responsibilities of investigators, institutional review boards (IRBs), and others involved in emergency research, FDA has released draft guidelines that spell out each group's responsibilities. The agency is now accepting comments on the document (www.fda.gov/OHRMS/DOCKETS/98fr/06d-0331-gdl0001.pdf) and will hold an 11 October public meeting on the subject. One concern for FDA is that some terms that guide emergency research, such as "life-threatening," may be defined differently by different people. In its proposal, the agency explains that "life-threatening" includes nonfatal risks, noting that emergency research on, say, victims of stroke or head injury could explore a treatment's ability to prevent disability as well as death.

Emergency research came under scrutiny earlier this year after *The Wall Street Journal* described a blood-substitute trial in trauma patients unable to consent, in which some suffered heart attacks. FDA officials said in a conference call last week that its review had already been under way and was unrelated to the blood-substitute flap. "It's taken time for us to develop and gather a sizable body of data on how this regulation has actually worked," said Sara Goldkind, an FDA bioethicist. The agency, she notes, has received roughly 60 applications for emergency research that allows for exceptions to informed consent and so far has approved about 20.

Physicians who perform such trials ▶

STEM CELL RESEARCH

# Scientists Object to Massachusetts Rules

Massachusetts stem cell researchers thought they were home free last year when the state legislature, overriding a veto by Republican Governor Mitt Romney, sanctioned research using human embryonic stem (hES) cells. But newly adopted final regulations to implement that legislation would cut off what some argue is an important potential avenue of stem cell research.

In May 2005, state lawmakers passed a measure that explicitly permits scientists to do things that federally funded researchers cannot—derive new lines of hES cells, including disease-specific lines produced using somatic cell nuclear transfer (SCNT), otherwise known as research cloning. The law allows ES cell lines to be produced from spare embryos left over after in vitro fertilization but prohibits the "donation" of embryos created just for research via IVF. Violating that provision, added to satisfy those who worry about "embryo farms," is punishable by up to 5 years in jail and a $100,000 fine. But the wording does not forbid scientists from working with such embryos if they weren't made in Massachusetts.

Romney tried unsuccessfully to amend the bill so that "use" of any such embryos in research would also be illegal. After the Democrat-controlled legislature overrode his veto, the state Department of Public Health trumped the lawmakers by inserting the wording Romney wanted into the regulations. "The prohibition on the creation of embryos [by fertilization] solely for use in research is implicit in the language" of the law, contends the Public Health Council, the nine-member body that makes the regulations. "[W]here the primary purpose is research, only the asexual creation of an embryo is permitted."

When the proposed regulation was presented in May, eight Boston medical institutions argued that it would "give the force of law to a provision the legislature specifically rejected." Scientists from those institutions reiterated their concerns last week when the final rules appeared. Harvard stem cell researcher Kevin Eggan says the regulation would prevent Massachusetts scientists from using cell lines derived in other states if they came from embryos created for research purposes. He stresses that it's important to preserve this option as an alternative to SCNT—which has not yet been proven—for creating disease-specific cell lines.

**More options.** Harvard's Kevin Eggan says purpose-bred embryos may be needed if nuclear transfer doesn't work for creating disease-specific cell lines.

But some scientists question the rule's impact on research. "I don't see it as a problem," says stem cell researcher Evan Snyder of the Burnham Institute in San Diego, California. "Most scientists agree that you don't want to make embryos specifically for research," he says, because it appears to be "ethically dicey."

The lawmakers are prepared to reassert their authority, starting with a hearing later this month. The leading gubernatorial candidates in the fall election (Romney is not running for reelection) support stem cell research, suggesting that the political winds are also favorable for a revision. **–CONSTANCE HOLDEN**

CREDITS (TOP TO BOTTOM): PHOTOS.COM; JUSTIN IDE/HARVARD

agree that the existing rules can be bewildering. "There's been a lot of anxiety and some confusion … about these regulations and how to apply them," says Lynne Richardson, an emergency-medicine specialist at Mount Sinai School of Medicine in New York City. For example, the dozens of IRBs overseeing a nationwide defibrillator study in which Richardson was involved required wildly different levels of community consultation.

Graham Nichol, who directs the University of Washington Harborview Center for Prehospital Emergency Care in Seattle, believes that confusion over the current rules has discouraged appropriate emergency research and, by making it difficult to follow up with subjects after treatment, sometimes failed to protect patients. The number of published cardiac-arrest trials in the United States has decreased since the rules were implemented while the number of non-U.S. studies grew, he found.

Will the new draft guidelines help? "I'm not sure they're any better," says Nichol, calling them still "too full of nuance." But, says Richardson, the new guidelines are clearly "an attempt to make sure that all of the research that actually qualifies in FDA's view" can go forward. **–JENNIFER COUZIN**

SCIENCE FUNDING

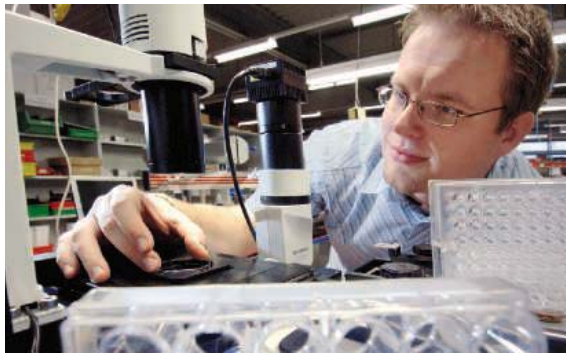
# Germany Launches a High-Tech Initiative

**BERLIN**—If Bill Gates had tried to start Microsoft from his father's garage in Germany, it never would have worked, says Holger Frommann of the German Venture Captial Association in Berlin. Among other things, he says, the government would have said that the garage didn't have enough windows to be a proper working environment. And whereas high-tech start-ups need less than a week to register in the U.S. or the U.K., he says, in Germany it can take much longer to complete the paperwork.

The German government says it wants to make it easier for a German Bill Gates to translate research discoveries into products; to this end, it is increasing support for programs that help spin scientific findings into commercial ventures. In a wide-ranging "high-tech strategy" announced last week, the government says it will spend €14.6 billion ($19 billion) in the next 3 years to boost technology-based research and enterprises, including about €6 billion in new funding.

The government wants to "ignite ideas," with a combination of new programs, funding schemes, and legislation, according to a multiagency strategy that Chancellor Angela Merkel and Research Minister Annette Schavan announced on 30 August. Researchers who collaborate with small- and midsized companies, for example, will qualify for a 25% funding premium from the government, up to €100,000. The government says it wants to change the tax law to encourage venture capitalists to invest in start-up companies. And the agriculture ministry has promised a new law governing genetically modified plants that should clear the way for more field trials.

The plan also includes several new funding schemes. Some €80 million would back technologies aimed at preventing terrorist attacks and disaster prevention and response, and €800 million would foster health and medical technologies, including new support for clinical research and teaching hospitals. The two largest investments are €3.65 billion for aerospace research, including satellite communication and navigation systems, and €2 billion for energy technologies, including biofuels and nuclear energy.



**Lowering barriers.** The German government wants to make it easier to turn research results into profits.

Tax breaks for start-up companies could be especially important, says Hans-Jürgen Klockner of the German Association of Biotechnology in Frankfurt. German scientists and industry leaders have long sought venture capital tax laws more in line with those of France and the United Kingdom. The details will be ironed out this fall in talks with the finance ministry. **–GRETCHEN VOGEL**

CREDIT: JAN-PETER KASPER/EPA/CORBIS

## Vatican Policy: Not Evolving

Don't look for a big change any time soon in the Catholic Church's views on evolution. Although supporters of evolution had feared that the Pope would embrace so-called intelligent design, Pope Benedict XVI gave no sign at a gathering last week as to how he thought the topic should be taught.

The pope said little during the meeting, which included his former theology Ph.D. students and a small group of experts near Rome. Peter Schuster, a chemist at the University of Vienna and president of the Austrian Academy of Sciences, attended the meeting and gave a lecture on evolutionary theory. "The pope … listened to my talk very carefully and asked very good questions at the end," he says. And the Church's most outspoken proponent of intelligent design, Cardinal Schönborn, seemed to distance himself from the theory. **–JOHN BOHANNAN**

## EPA Urged to Tighten Smog Rules

A scientific advisory board plans this month to recommend that the U.S. Environmental Protection Agency (EPA) lower the allowable level of ground-level ozone, which aggravates asthma and other health problems. The current legal limit is 0.08 parts per million (ppm). EPA scientists concluded earlier this year that the agency should either retain its current standard or tighten it to 0.07 ppm.

A majority of the 23 members of the Clean Air Scientific Advisory Committee (CASAC) said in a meeting last month that the standard should be 0.070 ppm, while two called for a slightly higher level. Once the panel's official recommendation arrives, EPA has until March to set final standards. CASAC "is throwing down the gauntlet," says Frank O'Donnell of the nonprofit Clean Air Watch in Washington, D.C. "Is it about science or politics?"

**–ERIK STOKSTAD**

## A Bang-Up Job

The European Space Agency's diminutive Smart-1 probe ended its 3-year technology mission this week with a lunar crash landing after successfully testing a propulsion system that fires out xenon ions. "Smart-1 has left a legacy of technology and scientific excellence," said mission scientist Bernard Foing. A camera and two spectrometers on board yielded information on the lunar surface including data on calcium, which could help scientists pinpoint the age of the moon. Researchers also say the crash itself could give clues about how craters form.

**–DANIEL CLERY**

UNIVERSITY FUNDING

# Academic Earmarks: The Money Schools Love to Hate

An unusual query from a "pork-busting" U.S. senator has revealed an uneasy ambivalence among university presidents toward academic earmarks. Their answers suggest that, like it or not, such directed spending on research is now part of the fabric of higher education.

On 27 July, Senator Tom Coburn (R–OK) asked 110 U.S. universities to describe any federal research dollars obtained in the past 6 years through the good graces of their congressional delegations rather than via a competitive review. He also wanted to know which universities have hired lobbyists to help obtain earmarks and the impact of the found money on their campuses and on science.

Coburn, who chairs a Senate financial management subcommittee, calls research earmarks, which have grown into a multibillion-dollar-a-year phenomenon (see graphic), "a gateway drug to overspending." His six-question letter set off a month-long frenzy of meetings and conference calls among vice presidents for sponsored research, directors of federal relations, professional associations, and lobbyists to figure out how, and whether, to respond. Only 14 schools met Coburn's 1 September deadline, although a few told him they needed more time.

Respondents, which included major research universities and leading recipients of federal earmarks, offered varying views of earmarking. But even those who said they abhor the practice acknowledged occasional dalliances. Cornell University President David Skorton, for example, cited "a long-standing and well-documented policy of not pursuing or accepting earmarks from federal agencies that award funds on a competitive basis" before acknowledging, two paragraphs later, that "Cornell makes two exceptions to this policy." The biggest is earmarked funds from the Department of Agriculture's cooperative research and extension service, which provides about 1.5% of the university's $381 million federal research budget. "They've worked on the basis of earmarks since 1865," explains Robert Richardson, Cornell's vice provost for research, about a program he says is essential to fulfilling Cornell's role as a land-grant college.

The University of Michigan shares Cornell's distaste for pork, says Stephen Forrest, vice president for research, although his reply to Coburn notes that Michigan last year received three earmarks totaling $5.3 million. In fact, the university has adopted a formal application process—much like a grant proposal in its length and complexity—for faculty members who think their idea deserves to be one of the school's "rare exceptions" (www.research.umich.edu/policies/earmarkpolicy.html).

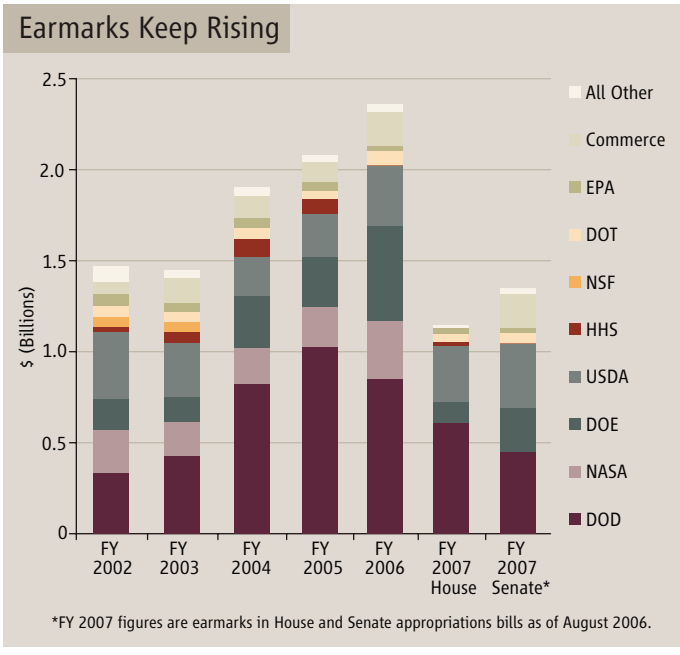
Some universities see earmarks as a way to simultaneously move up the academic food chain and strengthen the local economy. "The direct appropriations that the Kentucky delegation works hard to acquire for the university are an important part of UK's federal funded projects," writes Lee Todd Jr., president of the University of Kentucky, who notes that his school has received "over 100 [since 2000] worth a total of $120 million." Wendy Baldwin, U.K. vice president for research and the former head of extramural research at the National Institutes of Health, explains that earmarks "can help us to get into the top 20" recipients of federally funded research by public universities. The university closely monitors how the money is spent, she says, adding that "we expect people to advance based on this boost."

Not every institution is as comfortable as Kentucky is in speaking openly of its appetite for earmarks. University of Missouri President Elson Floyd, for example, provided the same answer to two of Coburn's questions, saying curtly that "all specific objectives and goals [for the research funded by the earmark] are outlined by the granting Federal agency… and specific measures of success are determined by [those] specific goals and objectives." And Floyd gave one-word answers—no, yes, and yes—when asked whether Missouri has a policy on earmarks, hires lobbyists to snare them, and thinks they are beneficial to the school. (In an increasingly common practice among universities, Missouri retains a Washington lobbyist, Julie Dammann, former chief of staff to Missouri's senior senator, Republican Kit Bond, well-known for his earmarking prowess.)

John Hart, Coburn's communications director, says his boss blames his legislative colleagues more than the academic community for what is happening. "The earmark process doesn't help universities so much as it helps lobbyists and Congress," says Hart, who notes that Coburn has held dozens of hearings on all manner of federal spending practices. "Because every time they get an earmark, the politicians can hold a press conference to claim credit."

Not surprisingly, Coburn's aggressive campaign has angered influential senators who are also heavyweight porkers. Senator Ted Stevens (R–AK), chair of the Senate Appropriations Committee and author of the notorious $225 million "bridge to nowhere" earmark for his state, has so far blocked Coburn's bid to create a publicly accessible database of Senate earmarks. And many legislators are said to be incensed that Coburn went over their heads in asking universities how they obtained specific earmarks.

Those tensions are a big reason that universities found Coburn's letter so troublesome. "The last thing you want to do," explains one university lobbyist, "is to get caught in the middle of a fight between two powerful senators." **–JEFFREY MERVIS**




**Research a la carte.** Congress has become increasingly fond of larding agency budgets with university research projects based in their districts.

SOURCE: AAAS 2006

**Courting disaster.** Coastal flooding is a likely impact of climate change, researchers tell the U.S. Supreme Court.

CREDIT: STEPHAN SAVOIA/AP PHOTO

CLIMATE SCIENCE

# U.S. Supreme Court Gets Arguments For EPA to Regulate $CO_2$

Can a 36-year-old U.S. law intended to reduce air pollution keep up with science? The U.S. Supreme Court will address the question this term in a case about whether greenhouse gases such as carbon dioxide should be regulated as pollutants. Several prominent climate researchers hope the court will also correct what they see as a distortion by a lower court and the federal government of the current state of the science.

First passed in 1970, the landmark Clean Air Act gave the new Environmental Protection Agency (EPA) the ability to tackle new pollutants as researchers discovered them. The law requires EPA to set vehicular emission standards for substances that could "reasonably be anticipated to endanger public health or welfare." But although the statute defines effects on "welfare" to include impacts on climate as well as on soils and water, the agency has used the act to regulate smog and other pollution from cars—not greenhouse emissions.

In 1999, as scientific evidence of climate change impacts accumulated, a Washington, D.C., nonprofit organization petitioned EPA to change its mind. EPA declined, and in 2003 a number of states and nonprofit groups sued. That case, *Massachusetts* v. *EPA*, is now before the Supreme Court, and last week 12 states and a number of cities and nonprofit groups filed their arguments.

The filing coincides with new state limits for industrial emissions passed by the California legislature last week. "We cannot do the job alone," said Ross C. "Rocky" Anderson, mayor of Salt Lake City, Utah, in a press briefing last week. EPA says it won't touch the issue because, among other things, "numerous areas of scientific uncertainty" surround climate change. Because greenhouse gases aren't pollutants, EPA officials assert, the agency doesn't have the authority to regulate them.

What's especially galling to a number of prominent climate scientists is the agency's use of a 2001 White House–requested report from the National Academies' National Research Council (NRC). It stressed the scientific consensus on climate change but noted that the "health consequences … are poorly understood." The report also cites the challenge of differentiating between anthropogenic climate change and "natural variability." Massachusetts and its allies believe that the appeals court erred in its July 2005 ruling that gave EPA broad discretion to avoid a rigorous scientific analysis of the harmful effects of carbon dioxide.

In a friend-of-the-court brief filed last week, a group of researchers says that the scientific evidence "is clearly sufficient" to support a "reasonable anticipation" of the risks of greenhouse gases. Both EPA and the appeals court "mischaracterized" the 2001 report by quoting from it selectively, they add. "We have the responsibility to correct when science is misrepresented," says Inez Fung, a University of California, Berkeley, climate researcher and one of six members of the 2001 climate panel who signed onto the brief. Panel chair Ralph Cicerone, now president of the National Academy of Sciences, declined to join the effort, a spokesperson said, because NRC reports "can and must stand on their own."

EPA, with allied states and industries, will file its arguments next month. Jay Austin, an attorney with the Environmental Law Institute in Washington, D.C., says that Massachusetts's reliance on the text of the 1970 law could play well with a majority of the justices, who are expected to rule before their term ends in June. **–ELI KINTISCH**

## SCIENCESCOPE

### Asian Alliance

**NEW DELHI**—India's science minister, Kapil Sibal, was in Beijing this week to ink a new accord that would pave the way for a high-powered Steering Committee on S&T. That body, chaired by Sibal and his Chinese counterpart Xu Guanhua, is expected to remove bureaucratic obstacles to cooperation in areas including genomics, weather forecasting, earthquake prediction, and nanotechnology. "We cannot lag behind China," says Sibal, who calls the steering committee a step in the right direction.

The first-ever visit of an Indian science minister to Beijing comes as Indian leaders express concern over China's burgeoning support for R&D. India today spends about $5 billion on R&D per year, amounting to 0.9 % of gross domestic product. In 2003, China spent about $85 billion, or 1.3% of its GDP, on R&D. **–PALLAVA BAGLA**

### Tomes on Genomes

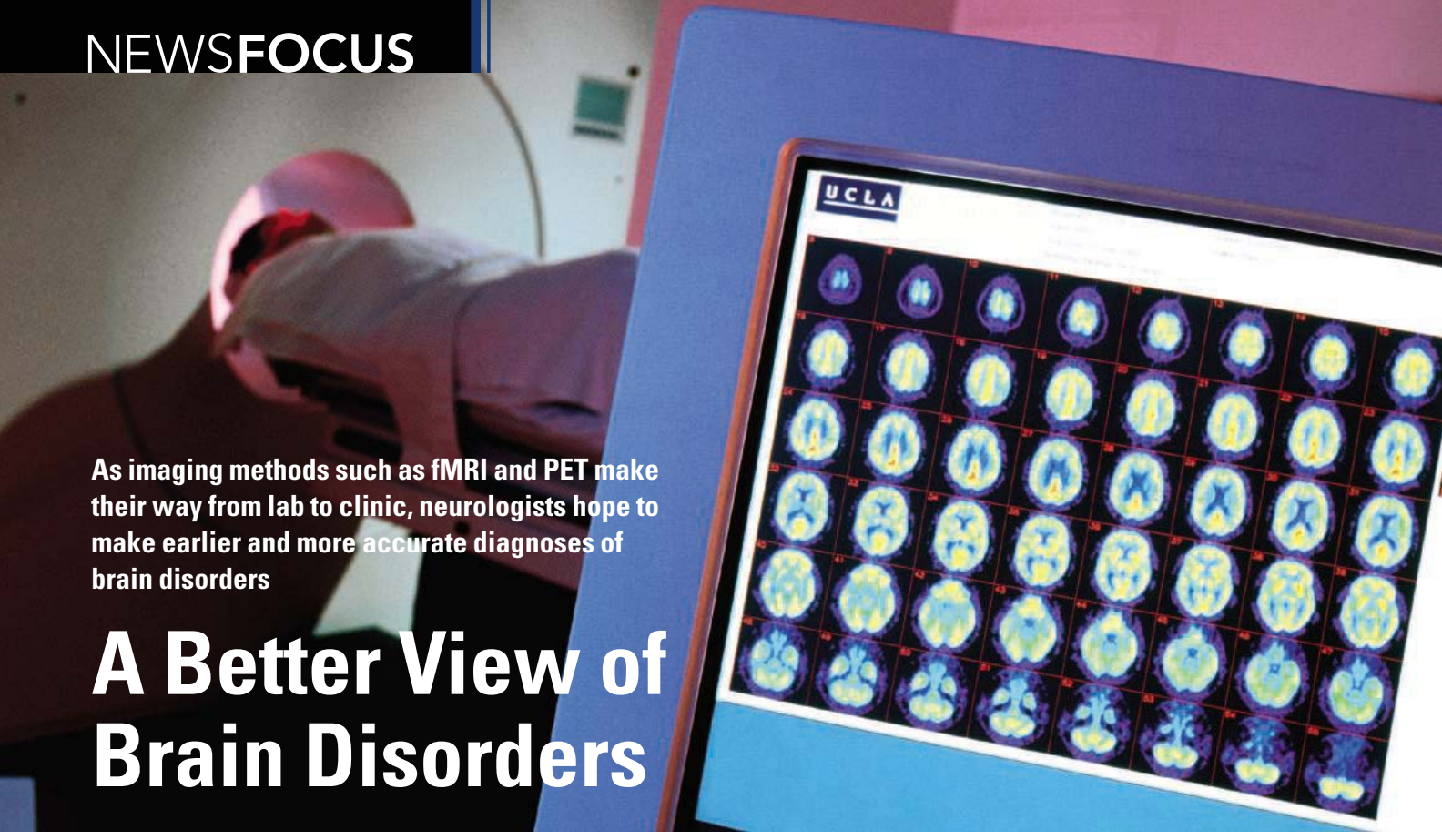
Already the home of GenBank, the global storehouse of genome data, the U.S. National Institutes of Health (NIH) now plans to create a free, central database for studies about links between genes and diseases such as cancer and diabetes. If adopted, NIH's new policy will urge NIH grantees conducting so-called genomewide association studies to share deidentified genetic and clinical data before publication. One provision that could prove controversial is NIH's desire to discourage researchers from patenting their initial data, which could slow the development of new drugs, warns Hakon Hakonarson of the Children's Hospital of Philadelphia. Comments are due by 31 October.

**–JOCELYN KAISER**

### KOMP Commences

The U.S. National Institutes of Health has chosen four centers for a $50 million effort to create knockouts in 10,000 mouse genes. The endeavor—dubbed the Knockout Mouse Project (KOMP)—is part of a global initiative to knock out every gene in the mouse genome (*Science*, 30 June, p. 1862).

Children's Hospital & Research Center Oakland in California will create the genetic material that the Wellcome Trust Sanger Institute in Hinxton, U.K., will use to knock out thousands of genes in embryonic stem cells. Researchers at the University of California, Davis, School of Veterinary Medicine will then create adult mice from these cells. Regeneron Pharmaceuticals, based in Tarrytown, New York, will perform all three steps. **–DAVID GRIMM**



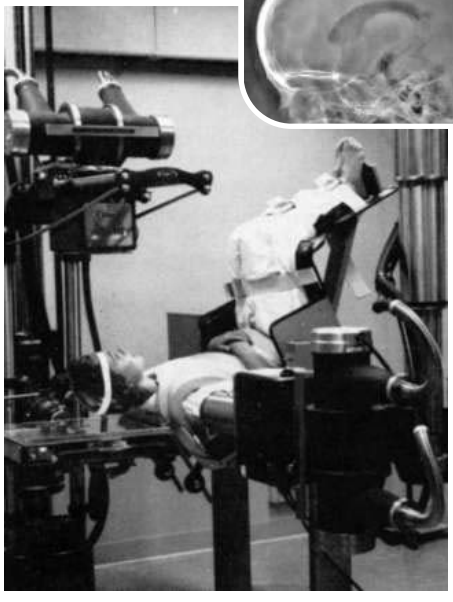

**As imaging methods such as fMRI and PET make their way from lab to clinic, neurologists hope to make earlier and more accurate diagnoses of brain disorders**

# A Better View of Brain Disorders

**IT WASN'T SO LONG AGO THAT TURNING** a patient upside down was the state of the art in clinical brain imaging. The technique, called pneumoencephalography, involved injecting air bubbles into the fluid surrounding the spinal cord and strapping the patient into a rotatable chair. As the chair swiveled, the bubbles floated upward and moved along the surface of the brain, allowing a series of x-ray images to better distinguish its contours. "You put the x-ray images together in your mind's eye, and you'd get a picture of the brain," recalls Marcus Raichle, a neurologist at Washington University in St. Louis, Missouri, who learned the method in the late 1960s. Pneumoencephalography helped neurologists find tumors and diagnose other problems that altered the gross anatomy of the brain. But the films were hard to interpret, Raichle says, and the procedure gave patients a nasty headache.

The advent of x-ray computed tomography scans in the early 1970s made pneumoencephalography obsolete almost overnight. When magnetic resonance imaging (MRI) came into clinical use in the early 1980s, it gave neurologists even more detailed snapshots of the brain's structure. But these techniques have shortcomings as well. Unlike, say, a femur, the fitness of the brain is hard to assess from still pictures.

Slowly but surely, a new generation of brain-imaging methods is finding its way from research labs into the clinic—and these techniques are offering physicians a much more dynamic look into the brain. Functional MRI (fMRI), a method used since the early 1990s to infer brain activity in studies of human cognition, now helps neurosurgeons map patients' brains before surgery, and a report on page 1402 raises the possibility of using fMRI to determine whether a patient in a vegetative state has conscious thought.



**Old school.** Pneumoencephalography was unpleasant for patients and produced fuzzy x-ray images of the brain (*inset*).

Positron emission tomography (PET), another standard tool of cognitive neuroscientists, also has medical promise. Clinicians already use PET to distinguish Alzheimer's disease from other types of dementia, and they are investigating ways to use PET to diagnose Alzheimer's and other diseases before symptoms appear—and before substantial structural damage to the brain has occurred. Some scientists even envision a day when real-time images of a patient's neural activity will provide a treatment for chronic pain or guide therapy sessions for psychiatric disorders.

Obstacles remain, even for developing routine diagnostic applications, but many experts say clinical uses of these brain-research tools are long overdue. "There's no question it's the future of my field," says John Ulmer, a radiologist at the Medical College of Wisconsin in Milwaukee and president of the American Society of Functional Neuroradiology (ASFNR), a group founded in 2004 to promote clinical applications of brain-imaging tools such as fMRI and PET. "It's not going to revolutionize the treatment of brain diseases with one broad stroke, but it's entering the clinical realm gradually, and it's going to continue to grow."

CREDITS (TOP TO BOTTOM): MARK HARMEL/GETTY IMAGES; TAVERAS AND WOOD, *DIAGNOSTIC NEURORADIOLOGY*, WILLIAMS AND WILKINS CO. (1964)

**Inside look.** Long used in research, PET brain imaging is gaining a foothold in neurological practice.

### A "spectacular result"

The case study reported in this week's issue of *Science* (see related Perspective on p. 1395) hints at how measures of neural activity can provide a dramatically different picture of the brain than that gleaned from now-routine structural MRI scans. Adrian Owen, a neuroscientist at the Medical Research Council Cognition and Brain Sciences Unit in Cambridge, U.K., and his team used fMRI to examine brain function in a young woman who sustained severe head injuries last year in a traffic accident. Five months after the accident, she was unresponsive, unable to communicate, and met the clinical criteria for vegetative state.

However, fMRI scans showed that language-processing regions of her brain became active when words were spoken to her but not when she was exposed to nonspeech sounds. Sentences containing ambiguous words such as "creek/creak" activated additional language regions, as they do in healthy people. These findings indicated that she retained some ability to process language, Owen says.

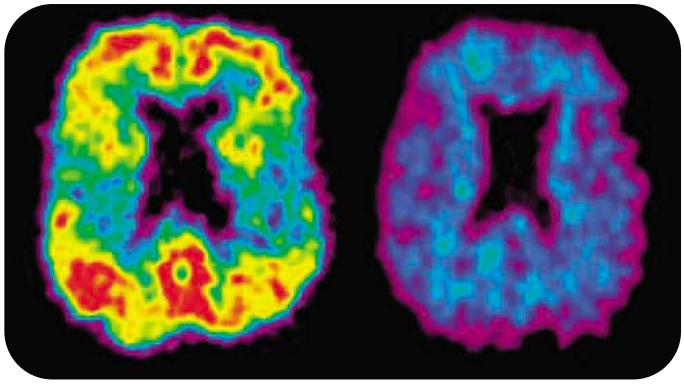
In another test, the researchers instructed the woman to picture herself playing tennis or walking through her house. In healthy people, imagining each activity activates a different set of brain areas involved in planning movements. The patient's fMRI scans showed an identical pattern—clear evidence, Owen and colleagues say, that she made a conscious decision to follow their instructions.

Although some researchers aren't convinced Owen's team has cinched the case for consciousness in this woman, most agree that the fMRI scans reveal evidence of cognition that could not have been anticipated from standard MRI scans. "It's a spectacular result," says Nicholas Schiff, a neurologist at Columbia University.

Owen hopes to build on this work to develop a battery of fMRI tests for measuring cognitive functions in brain-damaged patients who are unable to communicate. He says this approach might someday be used to customize a patient's rehabilitation. For instance, if a patient's fMRI scans revealed an incapacitated visual system but a working auditory system, therapists could employ speech and sound. It's a wonderful idea, says Schiff, but a "staggering" amount of work is needed to make it happen.

Yet fMRI has already made some clinical inroads, most notably in presurgical planning. For example, patients with tumors in the left frontal lobe of the brain present an especially tricky challenge for neurosurgeons trying to remove the cancer without destroying nearby brain tissue that controls speech and movement. Ulmer and his colleagues have been using fMRI to map out the brain regions responsible for these functions in presurgical patients, and they've recently added on an MRI method called diffusion tensor imaging (DTI) to map the tracts of axons conveying information from one brain region to another. Surgeons use this road map to determine how to reach a tumor and how much tissue to remove, Ulmer says. "We've seen a fivefold decrease in neurological complications with [combined fMRI and DTI] mapping for left frontal lobe tumors at our institution," he says.

Researchers and clinicians are still experimenting with DTI, and most hospitals don't have the equipment and expertise to use it.



**Signs of trouble.** In PET scans, PIB lights up regions of β-amyloid accumulation (red-yellow) in an Alzheimer's patient (*left*) but not in a healthy control (*right*).

CREDIT: UNIVERSITY OF PITTSBURGH AMYLOID IMAGING GROUP

More physicians have already embraced fMRI. In 2004, 30% of neuroradiologists responding to an ASFNR survey reported that their institutions used fMRI for presurgical planning; with nearly double that number expecting to use it.

Scientists are also excited about using fMRI in the early diagnosis of Alzheimer's disease. Although there are currently no drugs capable of slowing the disease's rampage through the brain, early diagnosis will be key if such drugs are found. Otherwise, any intervention may be too late to reverse the damage done.

In 2004, Michael Greicius, a neurologist at Stanford University School of Medicine in Palo Alto, California, and colleagues reported in the *Proceedings of the National Academy of Sciences* (*PNAS*) that they'd used fMRI to distinguish people with mild Alzheimer's disease from healthy elderly people. Alzheimer's patients at rest had less activity in a "default network" of brain regions, first identified by Raichle and colleagues, that includes certain regions of the cerebral cortex and the hippocampus, a crucial memory region. Such changes probably reflect a long-term decline in cellular metabolism caused by the disease, Greicius says. Although other researchers have argued that using fMRI to monitor brain activity in subjects engaged in memory tests should be the most sensitive way to pick up early signs of Alzheimer's disease, Greicius fears that smaller hospitals may not have the expertise to do task-activated fMRI. His approach—if it proves its merit in larger trials—would be far easier to use. "It's the sort of thing that could be done at a community hospital, where a technician presses a button and says, 'Keep your eyes closed,' and the software does the rest."

Scott Small, a neurologist at Columbia University, is taking what he thinks is a more targeted approach to picking up early signs of Alzheimer's disease. Like Greicius, he's using fMRI to look for long-term changes in brain metabolism rather than for short-term changes in brain activity evoked by a task. But instead of using BOLD fMRI, which measures blood oxygenation and is widely used by researchers to infer neural activity, Small has been working to refine a variant of fMRI that measures a different indicator of metabolic activity, blood volume.

There's an emerging consensus that Alzheimer's disease strikes the hippocampus first and afflicts some parts of the structure before others, Small says. The blood-flow method provides better spatial resolution—enough to distinguish hippocampal subregions—and is easier to interpret than BOLD fMRI, Small says. His studies on animal models of Alzheimer's disease and preliminary work with people suggest that the earliest detectable sign of the disease is reduced metabolism in the entorhinal cortex, a region closely connected to the hippocampus. Small and colleagues at Columbia now have a grant from the National Institute on Aging to evaluate the diagnostic potential of the method in up to 1000 elderly people.

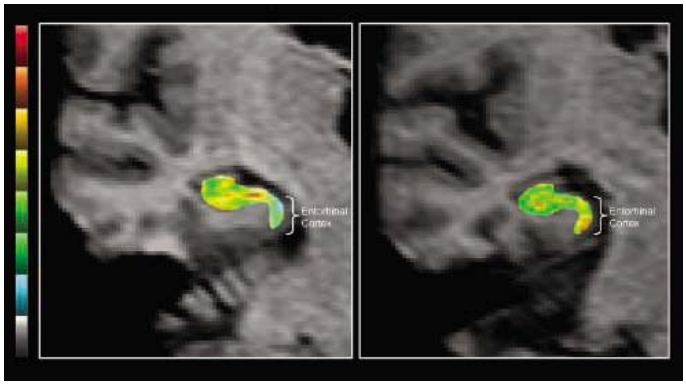
### Neurologists' PET

In the Alzheimer's arena, fMRI is a step or two behind PET. So-called FDG-PET, which measures glucose uptake in the brain, another metabolic indicator and proxy for

neural activity, has been used in recent years to distinguish Alzheimer's disease (which reduces metabolism in temporal lobe structures such as the hippocampus) from frontotemporal dementia (which reduces metabolism in the frontal lobes) in people with signs of dementia. It's become more popular since Medicare began reimbursing doctors for the procedure in 2004.

FDG-PET has also shown promise for detecting Alzheimer's disease before symptoms appear. In a study reported in *PNAS* in 2001, a team led by Mony de Leon, a neurologist at New York University, used FDG-PET to monitor glucose metabolism in the brains of 48 healthy elderly volunteers. Three years after those initial scans, 11 of the volunteers had developed moderate cognitive impairments and one had developed Alzheimer's disease. Reduced metabolism in the entorhinal cortex during the initial scanning session was the measure that best predicted which people experienced a subsequent decline, de Leon and colleagues reported.

His team has recently completed a study of a larger group of elderly people followed for longer periods of time. "With FDG-PET, we can pick up changes [in the brain] 9 years before the onset of symptoms," says de Leon. He adds that work from his group and others suggests that maximizing the sensitivity and accuracy of diagnostic tests will require combining FDG-PET with other biomarkers, such as levels of Alzheimer's-related compounds like β amyloid and tau in the cerebrospinal fluid. A more comprehensive evaluation of FDG-PET's diagnostic promise should come from the 5-year, $60 million Alzheimer's Disease Neuroimaging Initiative (ADNI), a federally funded longitudinal study of 800 elderly people, half of whom will receive an FDG-PET scan.




**Blood loss.** Less blood volume (cooler colors) in the entorhinal cortex distinguishes a patient with early Alzheimer's disease (*left*) from a healthy elderly person (*right*).

ADNI will also investigate another potential use of PET: imaging β amyloid, the main ingredient in the β-amyloid plaques that are a defining characteristic Alzheimer's disease. In 2002, researchers hailed the long-awaited discovery of a radioactive compound that makes it possible to see β amyloid in the brains of living people (*Science*, 2 August 2002, p. 752). Several pharmaceutical companies are already using this compound, called PIB, in clinical trials to monitor the effectiveness of candidate Alzheimer's drugs aimed at reducing β-amyloid buildup in the brain, says PIB co-inventor William Klunk, a neurologist at the University of Pittsburgh in Pennsylvania. In July, the Alzheimer's Association announced a $2.1 million grant that will enable ADNI-funded researchers to incorporate PIB PET scans into their studies to evaluate the method as a diagnostic test for Alzheimer's disease.

The original version of PIB utilizes a radioactive isotope—carbon-11—with a half-life of just 20 minutes, limiting its use to hospitals with easy access to a cyclotron. Klunk, in partnership with GE Healthcare, has recently developed a version of PIB based on fluorine-18, which has a far more convenient 120-minute half-life. The first research studies with F-18 PIB in humans should be under way by the end of this year, Klunk says.

Several other PET-compatible β-amyloid-imaging compounds are under investigation around the country. "These are coming fast and furious," says Kenneth Marek, a neurologist and president of the Institute for Neurodegenerative Disorders, a nonprofit research institute in New Haven, Connecticut.

PET markers are also in the works for Parkinson's disease—and one is already in clinical use in Europe. A marker called DaTSCAN, also developed by GE Healthcare, uses radioactive iodine to label dopamine transporters, proteins in nerve terminals that recycle the neurotransmitter dopamine after it's released into the synapse. Such methods provide a general indicator of whether the dopamine system, which breaks down in Parkinson's patients, is working properly, Marek says, and in principle they should be able to spot trouble before a clinician can. "By the time you've developed symptoms, you've probably lost 50% of these dopamine transporters," he says.

Marek and colleagues have investigated another compound that labels dopamine transporters, β-CIT. In pilot studies using single-photon-emission computed tomography, a method similar to PET, it showed promise for distinguishing Parkinson's disease from other movement disorders. In a group of 35 suspected Parkinson's patients referred by a community neurologist to a movement-disorders specialist, the imaging results with β-CIT agreed with the patients' ultimate diagnosis more than 90% of the time—an improvement over the 75% accuracy of the initial diagnosis made by the referring doctors, the researchers reported in 2004 in the *Archives of Neurology*.


**Burning pain.** As they seek to minimize computer-generated flames, chronic pain patients in an fMRI machine are actually trying to quell neural activity in pain-processing regions of their brains (*right*).

CREDITS (TOP TO BOTTOM): SCOTT A. SMALL/COLUMBIA UNIVERSITY; CHRISTOPHER DECHARMS/OMNEURON INC.

### Extinguishing pain's flame

Some researchers argue that the clinical uses of PET and fMRI won't be limited to diagnosing brain disorders. In the 20 December 2005 *PNAS*, neuroscientists reported using fMRI to teach people with chronic pain to monitor and control their own brain activity—a high-tech version of biofeedback. The research team included scientists from Stanford and the Massachusetts Institute of Technology and was led by Christopher deCharms, a neuroscientist and president of Omneuron, a start-up company in Menlo Park, California.

Each patient slid into an fMRI scanner and watched a computer-generated flame flickering on a monitor. The intensity of the flame reflected, with a few seconds' delay, neural activity picked up by the scanner in the patient's right anterior cingulate cortex, a region implicated in pain perception. The patients who best learned to minimize the flame reported the greatest reduction of pain symptoms immediately after the session. Another group of patients whose flames were fed by neural activity in their posterior cingulate cortex, an area not associated with pain processing, showed no such reduction.

"I thought this was enormously clever," says Raichle. Biofeedback has been tried previously for chronic pain, he says, but this is the first attempt to specifically target the brain regions that process pain. DeCharms's team is now doing a larger trial with weekly neurofeedback sessions for pain patients and following up to see how long the effect lasts.

Omneuron is also experimenting with real-time fMRI to assist psychotherapy. The firm's preliminary work has been in people with obsessive-compulsive disorder (OCD). Last year, at the annual meeting of the Organization for Human Brain Mapping, deCharms and colleagues described the method. Patients with OCD lie in the scanner, where they see the computer-generated flame, as well as a video link to their therapist, who sits in the control booth and also keeps an eye on the flame.

It's far too early to say whether the method will work. One of the central challenges, deCharms says, is determining the best brain areas to fuel the flames. Fortunately, he adds, functional neuroimaging methods such as fMRI have already provided many clues about what regions are involved in many psychiatric disorders. "The big question for us is, 'How can we take this nearly 20 years of research and turn it into clinical applications?' "

**–GREG MILLER**

CREDIT: R. STONE/*SCIENCE*

**Priceless.** Changbai's stunning vistas are drawing increasing numbers of tourists—and increasing pressures on the landscape.

ECOLOGY

# A Threatened Nature Reserve Breaks Down Asian Borders

**Chinese and Koreans share a love of Changbai Mountain, which straddles their border. Now that the area is under threat, the two sides may join hands to save it**

**CHANGBAISHAN NATURE RESERVE, CHINA—** To many Chinese, Changbai Mountain, whose jagged volcanic summit cups a crater lake on the border of North Korea, is the fatherland of Manchurian emperors who rose to power during the Qing Dynasty 4 centuries ago. Koreans, meanwhile, revere the iconic peak, which they call Paektu, as the birthplace of their culture and the nerve center of resistance to Japanese colonial rule in the 1930s and '40s. For scientists, Changbai is precious for another reason: It's a unique set of ecosystems under siege. Now, a new Chinese initiative aims to save it.

Changbaishan Nature Reserve, the largest protected temperate forest in the world, is home to endangered Siberian tigers and the last stands of virgin Korean pine-mixed hardwood on the planet. It's "one of the most spectacular and relatively undisturbed ranges in China," says Burton Barnes, a forest ecologist at the University of Michigan, Ann Arbor, who conducted research here in the 1980s and early '90s. But aggressive logging along the reserve's Chinese edge, and conversion to croplands on the Korean side, threaten to turn Changbai into "an oasis in a sea of clear-cutting," says Wang Shaoxian, director of the Jilin Changbai Mountain Academy of Sciences (JCMAS).

The reserve, roughly half the size of New York's Long Island, is also under increasing pressure from the inside. Chinese hot-spring resorts and Korean revolutionary museums on Changbai's flanks—the rugged, isolated terrain provided cover for the resistance—have transformed the reserve into a tourist mecca.

Hoping to counter these threats to the fragile ecosystems, the Chinese government this year designated Changbaishan, or "Perpetually White Mountain," as a major research initiative in its latest 5-year plan. It's pouring money into new facilities and projects, including a biodiversity survey and a study of how to better manage the Changbai ecosystems. The venerated mountain may also become a symbol of science transcending boundaries. Chinese and North Korean forest ecologists, who have had scant contact in recent years, are discussing the potential for collaborations at Changbai. From the vantage of local authorities, such cooperation "would be incredibly possible," says Ding Zhihui, deputy director of the Jilin Changbaishan Protection, Development, and Management Committee.

A research stint at Changbai has long

been a rite of passage for many Chinese researchers. Biologists, volcanologists, and meteorologists would winter at a cliff-hugging station with stunning views of Heaven Lake (in Korean, Lake Chon). "That time of year, it's like the North Pole here," says Dai Limin of the Institute of Applied Ecology in Shenyang. Temperatures can plunge below –40°C, and heavy snowfalls make the winding road up the peak impassable for months. Only in 2001 did the hardy winter crews finally yield to automated stations. Year-round observations, especially volcanic monitoring, are critical, says Wang. Changbai has been quiet since minor eruptions in 1597, 1688, and 1702. "It's due," Wang says. Chinese spas are deemed within striking distance of future lava flows.


**Bald spots.** Logging and clear-cutting for crops have broadened the mountain's bare patches, indicated in pink on this Landsat map.

Once the snow melts, the highlands teem with researchers. The Chinese Academy of Sciences (CAS) runs Changbai like a scientific boot camp, deploying an army of grad students and young researchers each summer. The ringlike ecological zones that change with altitude are a top draw. From the sky, the demarcation of forest types appears like a target, with the 2700-meter summit as the bull's-eye. "It's very unusual to have distinct ecological zones so easily observable in one area," says Wang. Outside the reserve, he notes, one would have to hopscotch thousands of kilometers to see all the forest zones on display at Changbai.

Barnes and other U.S. ecologists have made scientific pilgrimages to Changbai. "I was very impressed with the beauty and diversity of the area," says Mark Harmon of Oregon State University, Corvallis. "The buzz of the bees in the basswood trees was just amazing." With CAS colleagues, Hank Shugart of the University of Virginia, Charlottesville, is using Changbaishan as a test bed for modeling vegetation response to climate change across Eurasia.

But scientific affection has not translated into robust protection. "Although no tree is allowed to be logged within the reserve, biodiversity has been degraded due to other human activities," says Guofan Shao of Purdue University in West Lafayette, Indiana, who has mapped forest zones at Changbai. The most severe disturbances stem from the harvesting of two valuable commodities: ginseng roots and pine nuts. Wild ginseng is disappearing, so forest plots are cleared for ginseng plantations, causing erosion. And the removal of pine nuts impairs regeneration and forces animals such as the gray squirrel or the spotted nutcracker that feed on the nuts to find other food sources or die out. Local authorities, for the first time, have banned the collection of pine nuts in the reserve this year. As a result, says Shao, "they basically have to send people to guard the forest" during the summer months.

**Reaching out.** Wang Shaoxian hopes to work with North Korean scientists to restore Changbai's embattled ecosystems.

Jilin authorities created JCMAS earlier this year to strengthen and coordinate research in the reserve. Although Changbai boasts a panoply of life, including more than 2000 plant species, "there has never been a systematic survey," says Wang. Just such an initiative started last December and should be completed this autumn, he says. JCMAS plans to work with universities and CAS institutes to compile a DNA library of the reserve's flora and fauna. And Barnes says a comparison of Changbaishan's ecosystems with similar regions in Japan and eastern North America, "before further development renders them fragmented and domesticated, is of the highest international priority."

Such work would undergird an ambitious attempt to "balance the competing interests of tourism and environmental protection," Wang says. Down the road, he says, saving Changbai may mean extending the reserve's boundaries, which could require resettlement of villagers. Support for such a drastic measure might get a boost if UNESCO declares Changbai a World Heritage Site as expected in 2008, prompting a management and research policy vetted by international experts.

Chinese officials hope to kick off cooperation with North Korea in advance of World Heritage designation. "We're very interested in working with them to restore the ecosystems," says Wang. Since spring, he explains, the Chinese government has been providing "much more encouragement" for contacts with North Korean researchers. "The quality of their scientists is high," says Dai, who in 2002 visited North Korea's lakeshore research station, at the bottom of a zigzagging staircase hundreds of meters long that's visible from the Chinese side. And exploratory talks have begun on involving U.S. researchers in projects with North Korea and China. Barnes, for one, is eager. North Korea's forests "are one of the least well known to Western ecologists of any in the temperate zone," he says.

Wang should be in a position to host collaborations in autumn 2007, when JCMAS expects to complete construction of a new research building. In the meantime, he and his colleagues are happy to see a treasure of two cultures finally getting the scientific attention it deserves.

**–RICHARD STONE**

CREDITS (TOP TO BOTTOM) GUOFAN SHAO/PURDUE UNIVERSITY; R. STONE/*SCIENCE*

EVOLUTIONARY ECOLOGY

# Sex and the Single Killifish

**Males seem to be superfluous in one fish species but may come in handy when genetic diversity is needed**

Males—who needs them? Not the mangrove killifish. Made up primarily of hermaphrodites, the species reproduces just fine without the masculine touch. Yet male killifish do exist and can play a role in the species' survival, says John Avise, an evolutionary geneticist at the University of California (UC), Irvine. He and his colleagues have now shown that mangrove killifish are part of a select group of animals that use this unusual reproductive strategy, known as androdioecy.

This particular killifish "is the single species of any vertebrate that is doing this," says Stephen Weeks, an evolutionary ecologist at the University of Akron, Ohio.

Among androdioecious species, which include certain clam shrimp, barnacles, and nematodes, most individuals have a single gonad that produces both eggs and sperm, which meet internally before the eggs leave the body. But in each of these species, a few diehard males exist.

Until recently, evolutionary biologists considered androdioecy to be a transitory phase that occurs while a species, depending on its need for either genetic diversity or reproductive self-sufficiency, switches from two separate sexes to hermaphroditic, or vice versa. One reason is that "it's a high evolutionary hurdle" for males to persist among hermaphrodites, explains Loren Rieseberg, an ecologist at the University of British Columbia, Vancouver. "Males need twice the fertility of hermaphrodites."

Weeks has found that clam shrimp have no trouble jumping this hurdle, suggesting that for at least some species androdioecy is a viable, long-term solution. He recently added nine new species of clam shrimp to the list of androdioecious shrimp, for a total of 13. Moreover, the phylogeny and biogeography of these species indicate that this male-hermaphrodite strategy has lasted between 24 million and 180 million years, Weeks and his colleagues reported online in the 6 December 2005 *Proceedings of the Royal Society B*.

Avise is just beginning to piece together the story of the mangrove killifish. It lives in the muck around the roots of mangroves in the Caribbean and along the coasts of South Florida and northern South America, hanging out in crab burrows and dead logs. Self-fertilization by the hermaphrodites yields offspring that are virtual clones of the parent, which is why researchers once expected to see little genetic diversity among killifish at any particular location.

**Going it alone.** Neither the mangrove killifish (*bottom*) nor the clam shrimp (*top*) needs a male to reproduce.

But 15 years ago, ichthyologist Bruce Turner of Virginia Polytechnic Institute and State University in Blacksburg discovered that certain populations had unexpectedly high levels of genetic diversity. He proposed that these fish might have unusually high mutation rates or that fish immigrating from other populations were the source of this variation. "Turner had it wrong," says Avise.

Working with colleagues, including Mark Mackiewicz of the University of Georgia, Athens, Andrey Tatarenkov of UC Irvine, and Turner himself, Avise collected killifish from along the Florida coast and analyzed their DNA. The group focused on 35 markers, DNA sequences called microsatellites, along the genome. In each population, the researchers found some individuals whose microsatellites were virtually identical. But, as they reported online 5 July in the *Proceedings of the Royal Society B*, some samples contained a few individuals whose DNA differed at so many markers that it raised suspicions that there was a second parent somewhere in the picture.

As far back as the 1960s, ichthyologists had demonstrated that they could, in the lab, produce male mangrove killifish by keeping self-fertilized eggs cool, for instance, or by growing immature hermaphrodites at high temperature. But little was known about what conditions produced males in the wild.

Avise and his team found very few males among the killifish collected in Florida or the Bahamas. But when they repeated the study with fish from Belize, 10% to 20% of the catches were male. And DNA analyses revealed dramatic differences in diversity among killifish from the various locations. Those from any one spot in the Bahamas or Florida were genetically similar, whereas members of Belize populations varied in their genetic makeup about as much as would be expected had they been following the typical male-female reproductive strategy, the researchers reported in the 27 June *Proceedings of the National Academy of Sciences*. More recently, Avise's group has confirmed in lab experiments that these males mate with the hermaphrodites and produce viable young that spice up the genetic diversity. They will report these results in an upcoming issue of the *Journal of Heredity*.

The existence of androdioecy in species as different as killifish and shrimp indicates that "there must be underlying biological commonalities in the kinds of selection pressures … and the evolutionary responses involved," says Avise. Weeks and other researchers think this strategy has worked so well—and for so long—in clam shrimp because they live in ephemeral pools and often find themselves trapped in new places sans partners. The widespread distribution of killifish suggests that it, too, is a good colonizer and that hermaphroditism may facilitate that skill, Avise adds.

But David Bechler, an ichthyologist at Valdosta State University in Georgia, suspects that hermaphrodites won't always have the upper hand among these killifish. Both he and Avise agree that mangrove killifish were once a two-sex species. And although conditions now favor hermaphrodites, the high proportion of males in Belize suggests that the low genetic diversity is becoming a handicap. "What we are seeing is male evolution reoccurring," Bechler suggests.

**–ELIZABETH PENNISI**

CREDITS (TOP TO BOTTOM): NAIDA ZUCKER; WILLIAM P. DAVIS

BIOENGINEERING

# Artificial Arrays Could Help Submarines Make Like a Fish

**An interdisciplinary team has developed nanostructures that mimic how marine animals hunt, evade prey, and stay in the swim of things**

Listen. As you read, tiny hair cells in your inner ear amplify and convert sound waves into electrical signals that can alert you to the output of your iPod or the approach of a subway train. Similar structures on other animals, such as seal whiskers and the hairs on spider legs, help those organisms to track prey and evade predators. Now, engineers and biologists have developed the world's first functional artificial hair cell to mimic one of nature's most widespread and versatile data-collecting systems: the lateral lines of fish.

**Close-up.** Researchers modeled flow sensors on tiny hair cells found on fish such as this mottled sculpin.

In a paper published in an August issue of *EURASIP Journal on Applied Signal Processing*, engineer Chang Liu of the University of Illinois, Urbana-Champaign, describes how biologically inspired microstructures enable a model fish to locate and track a dipole source. Real fish use a linear swatch of hair cells on their sides, known as the lateral line, to coordinate group movements, avoid predators, and otherwise navigate. "I'm thrilled to see this," says Jeannette Yen, director of the Center for Biologically Inspired Design at the Georgia Institute of Technology in Atlanta. "It shows that we do understand the biological system well enough to make a mimic that works in a similar way."

Morley Stone, a former program manager at the U.S. Defense Advanced Research Projects Agency (DARPA), which funds Liu under a project called BioSenSE (Biological Sensory Structure Emulation), hopes that artificial hair cells might someday be used to navigate crewless underwater vehicles too small to be equipped with cameras. The hair cells would greatly expand underwater imaging capacities beyond those now generated by sonar or cameras, he notes. "When you look through a soda straw, it's hard to get an idea of what your world looks like," says Stone.

Like their analogs in real fish, Liu's hair cells work by measuring the movement of nearby water. Most commercial flow sensors measure the change in electrical resistance when flowing water cools a heated metal wire. Although Liu has also developed lateral-line arrays using more conventional "hot wire" technology, his hair sensors, by contrast, are activated by force. These are made using a standard microfabrication technique called photolithography to carve polymers into long, flexible, narrow strands about 500 to 700 micrometers long and 80 micrometers in diameter. The strands are rooted in a silicon base called a pedal, creating a minuscule lever. When the hairs are bent, the strain on the pedal causes a change in electrical potential that correlates to flow velocity.

Liu tested his lateral-line array by installing it in an artificial fish. The model was attached via a rod to an agile motion stage whose positioning was directed by signals received by the fish in response to a wriggling dipole source. Although Liu's array used only 16 hairs rather than the 100 usually found on real fish, the artificial fish was able to target and track the moving source.

The BioSenSE team includes biologists, neurologists, engineers, and mathematical modelers, all working to reverse-engineer nature's blueprint. "This is one of the largest international groups we've been able to pull together," says Stone. For example, Sheryl Coombs, a neurobiologist at Bowling Green State University in Ohio, has collected data on the spatial distribution of pressure along the lateral line of real fish to develop algorithms sensitive enough to process the wealth of information gleaned by Liu's sensors. That information was then validated by numerical simulations carried out by biologist-engineer Joseph Humphrey of the University of Virginia, Charlottesville, and applied to the programming efforts of Douglas Jones, an engineer at the University of Illinois, Urbana-Champaign. "It illustrates the best of this new set of collaborations between biologists and engineers," says Steven Vogel of Duke University in Durham, North Carolina, who studies biomechanics.

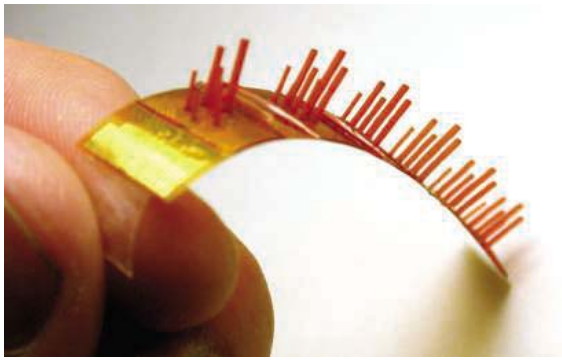
Coombs's experiments show that even blinded fish still orient themselves toward movement via a "map of touch" created by their sensory system. Abroad, zoologists Horst Bleckmann of the University of Bonn in Germany and Friedrich Barth of the University of Vienna in Austria are studying seals and spiders, respectively, for potential applications in both underwater tracking and airborne drones.

At Iowa State University in Ames, engineer Vladimir Tsukruk and his team used a synthetic hydrogel to mimic the soft cupula tissues surrounding fish hair cells that help relay information. The gel both protects the hairs against corrosion and makes them 10 times more sensitive. Liu's hair sensors can detect flows slower than 1 millimeter per second, half the rate of conventional

CREDITS: JAMES PARNELL; (INSET) TRISTAN ULA, PAUL PATTON, AND SHERYL COOMBS/BOWLING GREEN STATE UNIVERSITY

sensors. However, increasing the sensitivity of the sensor is a double-edged sword, says Liu, because of the added burden of filtering out unwanted noise. Scientists are using fish biology as a guide to tackle that problem as well, managing to mimic their hair cells' structural alignment that allows fish to weed out background noise.

Although the sensors were developed primarily to help guide small, robotic vehicles, Liu suggests that they could also assist submarines. For example, submarines now employ passive sonar to avoid giving away their position. But because that technology reads signals generated by noise, it cannot detect a stationary submarine or the subtle vortexes shed by large rocks. In addition, active sonar requires the emitted "ping" to travel away from the ship so that the feedback can be analyzed. That constraint creates a blind zone around the craft that makes subs vulnerable to sabotage by bomb-carrying divers, says Liu.



**The right bent.** The artificial hair cells are only 500 to 700 micrometers long and can be adapted to function as both vibration and tactile sensors.

Liu says that his array can eliminate that problem by detecting movement within a radius of about three to four times the length of the vessel, 200 meters or less for a full-sized submarine. Liu's hair cells are sensitive enough to detect both divers and large, unmoving bodies such as rock faces that are normally invisible in dark or murky conditions. Hair-cell sensors also have shown the potential to track other submarines based on wakes created minutes before, just as seals use their whiskers to track their prey. To turn those applications into reality, however, the artificial hair cells must be robust enough to withstand a marine environment.

Scientists can also imagine nonmilitary applications for the sensors. Changing the shape of the hair, Liu speculates, could yield vibration or tactile sensors in addition to flow sensors. Scaling up production could lower the cost of semiconductor sensors from $12 to $1 per unit, opening up markets as diverse as sneakers, MP3 players, and stress gauges in buildings in earthquake-prone areas.

Despite the many challenges, Stone predicts that DARPA will pick up the project for a second term beginning this fall. And if all goes well, someday hair cells might alert your iPod as well as your ear to the rumbling of an approaching subway train.

**–BRIAHNA GRAY**

CLIMATE SCIENCE

# Sea Animals Get Tagged for Double-Duty Research

**Elephant seals and other deep-diving species are providing an unexpected boost to a global oceanographic database**

Eight years ago, Dan Costa tagged nine elephant seals to learn how the sea mammals would respond to an expected El Niño event, a shift in a cold-water current in the Pacific. Sensors glued to the seals' backs were designed to record the depth at which they dived and the temperature of the water, while transmitters glued to their heads gave out their position. Once tagged, the giant pinipeds lumbered out from their rookery on Año Nuevo Island near Santa Cruz, California. Some went to the Aleutian Islands, others to the Gulf of Alaska, and a third group shot straight out West into the central Pacific.

After one season, the seals returned to Año Nuevo toting detailed records of 75,000 dives in the North Pacific. Costa, a biologist at the University of California, Santa Cruz, learned that the seals dive more frequently and deeper than previously thought—some 60 times a day, routinely as far down as 600 meters, and sometimes as deep as 2000 meters. In the last decade, tagging of this kind has given researchers increasingly sophisticated data from fishes, turtles, seals, and whales, revolutionizing our understanding of how they behave under the surface (*Science*, 11 August, p. 775).

**In depth.** The frequent, deep dives of California elephant seals provide a wealth of information about the ocean.

But in addition to the bounty of information on the animals' movements, their dives also pointed to a new method for scooping up hard-to-get information about the ocean that's useful for climate research. The method promises a wealth of physical data from the deep that will soon dwarf the amount gathered by ships and research buoys. And whereas the first wave of tagged elephant seals could only record depth and temperature, today's more sophisticated tags also capture salinity. "Different water masses have unique temperature and salinity signatures, and these can be used to trace the origin of the oceanic water in a given region," says Costa.

Researchers want to learn about temperatures and water density in the polar regions, for example, because they affect circulation and climate. James Hansen, chief of NASA's Goddard Institute for Space Studies in New York City, says that although researchers have collected data from the upper layers of most of the oceans, the polar regions are poorly covered. With support from ocean scientists, Costa and others are now tagging animals in these less explored areas, taking advantage of their ability to reach places where no machines can go.

### Seals as lab assistants

Looking over the collection of 75,000 depth profiles from elephant seals, Costa and his team thought the results might interest oceanographers. "But we had no idea what to do with the data, who to give it to, or how to prepare it," he recalls. That sum-

CREDITS (TOP TO BOTTOM): CHANG LIU AND JONATHAN ENGEL; DAN COSTA/UCSC/TOPP

mer, Costa presented the findings on El Niño's effects on elephant seals (surprisingly slight) at a meeting at the Scripps Institution of Oceanography in San Diego, California.

In the audience sat George Boehlert, then a lab chief at the U.S. National Oceanic and Atmospheric Administration (NOAA). "This was incredible data," recalls Boehlert, now head of Oregon State University's Hatfield Marine Science Center in Newport. "I was really surprised at the frequency of the dives and how deep these seals go." After the presentation, Boehlert told Costa he knew how to check the figures against existing data and, if they were accurate, how to enter them into a massive depot called the World Ocean Database (WOD).

**Big picture.** Dan Costa's team has been tagging elephant seals for 10 years at Año Nuevo Island near Santa Cruz.

NOAA had funded the database to hold records from ships and submarines. Later, it added data from its 2500 "Argo" buoys, which drift around the world at about 1000 meters below the ocean's surface, rising every 10 days to transmit temperature profiles. According to Sydney Levitus, the NOAA scientist who manages the database, each year Argo buoys provide 100,000 depth profiles, whereas other buoys, ships, and submarines provide about 140,000.

Back in 1998, Boehlert recalls, few oceanographers knew about animal electronic tags, and "among those who knew, there was a great deal of skepticism about the quality of the data." But the data proved reliable after being checked against profiles obtained by ships and satellites. So the 75,000 profiles from elephant seals were added to the ocean database. Boehlert, Costa, and Levitus also published a proof-of-concept paper in 2001 in the *Journal of Atmospheric and Oceanic Technology*. "You can't understand a climate system without knowing what's going on at depth," Levitus says. "So we want all the data we can get."

But the flow quickly dried up. What happened? After the California elephant seal study, Costa says, "we stumbled around trying to get funds to get tags, but we got nothing for years. We reused the tags we had," he adds, but "we had no money to pay someone to process the data." Although he and Levitus had shown the utility of the data for oceanography, that community has been slow to recognize its value—and to seek funding from the relevant federal agencies.

But that situation is changing, as interest in using tagging data for ocean research is on the rise. Since 2000, the Tagging of Pacific Pelagics (TOPP) program, funded mostly by private foundations, has been tagging 23 species in the Pacific Ocean. Seven of those species—the air-breathing ones that carry location transmitters—now produce about 1 million depth/temperature profiles a year. And TOPP hopes to format the data and deposit it in WOD within a year.

**Under the ice**

Two years ago, Costa and a team from Old Dominion University in Virginia won a 3-year, $800,000 grant from the National Science Foundation to join colleagues from France, the United Kingdom, and Australia in a program called Southern Elephant Seals as Oceanographic Sensors. The group is tagging 70 southern elephant seals, who then spend much of their time diving and feeding under the Antarctic pack ice. As they go about their business, the seals are gathering more than 10,000 profiles a year.

Antarctic data are critical for the study of ocean circulation, says Steve Rintoul, a U.S. oceanographer based at the Antarctic Climate and Ecosystems Cooperative Research Center in Hobart, Australia. Surface waters cool and become denser in the polar regions, sinking several kilometers to the ocean bottom. Warm water then flows in, creating the so-called thermohaline circulation. This process controls how much heat and carbon dioxide is stored by the ocean, influencing the rate of climate change. Climate models suggest that warming at the poles could slow down the circulation, driving further warming. But there are "almost no measurements," he says, because "subs aren't allowed … in this blind spot" and the Argo buoys can't transmit through the ice.

Meanwhile, Costa has turned over more than 1 million profiles—a decade of California elephant seal data—to Steven Bograd, an oceanographer with NOAA's Pacific Fisheries Environmental Laboratory in Pacific Grove, California. Bograd, another co–principal investigator for TOPP, is harmonizing and calibrating the data before comparing them with climatic events in the past decade, including two El Niño events. The goal, says Bograd, is to "better understand the mechanisms by which these climate signals impact the ecosystem."

So far, the most recent data from animal tags haven't gone into the ocean database, Costa says. "The reason it takes time is that we're coming up with much more precise and reliable methods of defining where the profiles were taken than we were in 1999," he says. "Five years ago, anything was valuable, but now it's compared to the Argo buoys, which are very precise."

How soon might these profiles be ready for the database? "We're working on it," Costa says. "I think we'll be able to turn over 2 years' worth of data, which is about 25,000 depth profiles, within 6 months." Oceanographers and climate researchers await the promised deluge.

**–CHRISTOPHER PALA**

Christopher Pala is a writer in Honolulu, Hawaii.

CREDIT: DAN COSTA/UCSC/TOPP

EDITED BY YUDHIJIT BHATTACHARJEE

## Pioneers

**LEADING BY EXAMPLE.** French astrophysicist Catherine Cesarsky last week became the first woman to be elected president of the 9800-member International Astronomical Union.

Cesarsky, 63, has been director general of the European Southern Observatory since 1999 and led the design and construction of the ISOCAM camera on board the Infrared Space Observatory of the European Space Agency. She previously headed basic research at the French Atomic Energy Commission.

Cesarsky welcomes the rising number of women graduating with Ph.D.s in astronomy but says that the challenge of juggling career and family keeps many from reaching their potential. She says she raised her two children, now adults, by using childcare and working at night. She willingly accepted some constraints on her career, she says, for the chance "to have a balanced life."

"She is very open, and she has a tremendous astronomical knowledge," says the union's new general secretary, Karel van der Hucht. Cesarsky says she will give all her support to the union's working group on women, which was created 3 years ago to monitor the status of female astronomers and promote gender equality and family-friendly measures.

## IN THE COURTS

**HITTING THE WALL.** A Florida State University (FSU) chemist who helped invent the blockbuster cancer drug Taxol has lost a court battle with his institution over how a portion of the royalties can be spent. But Robert Holton should be getting back an $11 million gift to the university from his foundation.

Holton, whose drug earned him and the Tallahassee school millions of dollars, pledged $18.5 million from a lab account held by the university toward a new building dedicated to his field, synthetic chemistry. He sued last year after FSU announced that the five-story building would be a general chemistry facility (*Science*, 18 November 2005, p. 1101).

In an oral ruling last week, Circuit Judge Janet Ferris threw out Holton's bid to prevent the university from spending the $18.5 million. But Ferris told FSU to give back the $11 million plus interest donated by the MDS Research Foundation established by Holton.

The foundation rejected the university's offer to return that amount in January because it also wanted the lab funds, says Michael Devine, the foundation's executive director. Holton may appeal the ruling, he says.

**NO BIAS.** An employment tribunal has ruled that the Roslin Institute in Midlothian, Scotland, and its former star scientist, Ian Wilmut, did not commit racial discrimination against a molecular biologist whom the institute fired 2 years ago. But the Edinburgh tribunal says the researcher, Prim Singh, was dismissed improperly.

Singh, 46, who now works at the Leibniz Center for Medicine and Biological Sciences in Borstel, Germany, accused Wilmut and the institute of dismissing his ideas because of his Asian heritage and sought $1.9 million in damages. The hearings exposed the dynamics of the team that created Dolly, the cloned sheep, and resurrected several old disputes over authorship and credit (*Science*, 17 March, p. 1539).

The tribunal ruled that Wilmut had been subjected to "wholly unjustified personal attacks by the claimant" but faulted the institute for not following due process in dismissing Singh. He could receive up to $114,000 in damages following a final hearing later this month.

## Three Q's >>

Nuclear physicist **Samuel Aronson** takes the helm of the U.S. Department of Energy's Brookhaven National Laboratory in Upton, New York, just as Congress prepares to restore funding for its Relativistic Heavy Ion Collider (RHIC) and provide money for the design of a proposed $700 million x-ray source.

**Q: How do you see the lab's mission evolving?**

Evolution is the right term. The science mission of the lab will not change, but there will likely be a rebalancing among the major thrusts. I expect RHIC and its upgrades to be active or under construction. The basic energy sciences' component will probably become a larger piece of our portfolio.

**Q: What are the challenges you foresee?**

We have several—and they are common to the entire national laboratory system—worker safety; security, including cybersecurity; and an aging infrastructure. We are making progress, but fiscal constraints cause us to move more slowly than I would like.

**Q: What science questions most intrigue you?**

My background is in experimental high-energy and molecular physics, and the fundamental questions addressed there continue to stimulate my personal interest. I have become particularly interested in the connection between nuclear and particle science in the laboratory and astrophysics and cosmology.

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# LETTERS

*edited by Etta Kavanagh*

## Declines in Funding of NIH R01 Research Grants

FOR MANY YEARS, THE NATIONAL CAUCUS OF BASIC BIOMEDICAL Science Chairs, an organization of medical school scientific faculty leaders, has followed U.S. NIH data on the likelihood of investigator-initiated unsolicited R01 research grant applications being funded (*1*–*5*). Research supported by these grants, which are the mainstay of research by medical school faculties and other research institutions, has permitted exploration of new approaches to understanding health and disease and development of therapies to treat illness.

We have collected data (*6*, *7*) on the fate of "unamended" (unsolicited) R01 applications. The unamended R01 represents the original application and does not consider resubmissions. NIH classifies R01 applications into type-1 (new) and type-2 (renewals). Revision and resubmission of initially rejected type-1 applications improve the likelihood of eventual funding by a factor of approximately two (*4*, *8*), with smaller increases for rejected type-2 grants. However, each revision of a rejected application delays by close to a year the time required before support can be approved and research initiated. For type-1 applicants, this is a slow, uncertain process that often leads to career reevaluation and change by otherwise successful professional contributors. For an ongoing and previously approved type-2 research activity, rejection casts major doubt on eventual continuation and frequently results in breaking up teams of highly trained personnel. Therefore, success rates for funding initial applications are of primary importance. It is encouraging that the review process itself may soon be accelerated.

The likelihood of funding type-1 and type-2 unamended, unsolicited applications reached a low-point in fiscal year (FY) 1993 and 1994: approximately 12% in each year for type-1 applications (*9*). For type-2 applications, success rates were 39 and 37%, respectively (*2*). Thereafter, success rates of unamended type-1 and type-2 R01 applications improved somewhat, peaking between FY 1999 and 2001 (*4*). Despite the doubling of the entire NIH budget between FY 1999 and FY 2003, success rates did not increase (*4*, *5*) (see table).

Since FY 2002, success rates have dropped steadily. In FY 2005, the decline was precipitous. Although the total number of applications has increased annually since FY 2002 (see table), not only success rates, but also total number of grants awarded and total dollars committed persistently decreased. For type-1 grants, an overall success rate of 9% has been calculated for FY 2005 (*10*). Peer review cannot discriminate among and accurately select only 1 of 11 meritorious applications. FY 2006 data are not yet available, but because the total NIH allocation for that period has been less than the biomedical inflation index, a trend toward further diminished support of R01 applications is evident.

Particularly surprising and regrettable is the continuing erosion in the allocation for total R01 annual funding of new unamended applications. This decreased from $510 million in FY 2002 for type-1 grants to $351 million in FY 2005 (see table). These dollar figures represent less than 1% of the entire NIH budget. Of similar concern is the 38% decrease in total number of unamended R01 applications awarded during this period for new applicants (type-1), even though submissions increased 24%. Major reductions are also evident in renewal applications for competing ongoing investigations (type-2).

This issue raises serious concerns about the present and future of U.S. biomedical science because the R01 grant is such an essential contributor to, and index of, scientific innovation. Recent discoveries have provided enormous new opportunities to better understand and treat disease, and we must take advantage of these breakthroughs. In addition, the country's economic future depends on U.S. leadership in providing new scientific and technical discoveries. Also, failure to provide adequate funds for biomedical research discourages the brightest young people from choosing scientific pursuits.

**Fate of unamended (unsolicited) R01 research grant applications (*6*)**

| Fiscal year | Number submitted | Number awarded | Total $ awarded (millions) | Success rate (%) |
|---|---|---|---|---|
| **Type-1 grants: new submissions** | | | | |
| 1999 | 8957 | 1761 | 456 | 19.7 |
| 2000 | 8626 | 1736 | 503 | 20.1 |
| 2001 | 8284 | 1590 | 501 | 19.2 |
| 2002 | 8560 | 1556 | 510 | 18.2 |
| 2003 | 9605 | 1477 | 493 | 15.4 |
| 2004 | 10624 | 1288 | 438 | 12.1 |
| 2005 | 10605 | 970 | 351 | 9.1 |
| **Type-2 grants: continuation (renewal) submissions** | | | | |
| 1999 | 3214 | 1772 | 554 | 55.1 |
| 2000 | 3233 | 1708 | 563 | 52.8 |
| 2001 | 3100 | 1637 | 583 | 52.8 |
| 2002 | 3153 | 1555 | 559 | 49.3 |
| 2003 | 3767 | 1697 | 627 | 45.0 |
| 2004 | 3773 | 1530 | 580 | 40.6 |
| 2005 | 3896 | 1262 | 496 | 32.4 |

**H. GEORGE MANDEL[1]*† AND ELLIOT S. VESELL[2]**

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6. Kindly provided by Office of the Director, Office of Reports and Analysis, Office of Extramural Research, NIH.

7. The R01 pool, as calculated by the NIH, includes a small number of R37 Merit Awards, but not Programs or Centers, and it separates out Program Announcements (PAs) and Requests for Applications (RFAs), which are not included in our calculations. Also excluded in our discussion are noncompeting renewals and the increasingly popular R21 grant mechanism, distinct from the R01, for short-term, introductory and exploratory research projects, with a limited budget (maximum total $275,000 over 2 years) and offered only by certain NIH Institutes. For these reasons, the R21 grant is not considered a substitute for the R01 long-term basic support of faculty.
8. See http://grants1nih.gov/grants/award/success.htm.
9. There also existed some R29 First Awards, now combined with the R01 pool, with slightly higher success rates.
10. Especially low success rates existed for the National Institute of Environmental Health Sciences (4%), the National Institute of Child Health and Human Development (5%), and the National Institute of Mental Health (5%).

## IRBs: Going Too Far or Not Far Enough?

IN THEIR EDITORIAL "MISSION CREEP IN THE IRB world" (9 June, p. 1441), C. K. Gunsalus and colleagues point out the frustration many have with an increasingly regulated Institutional Review Board (IRB) process that places all human subject research in a fish bowl. However, I see no evidence that the IRBs are neglecting their duties for thoughtful consideration of ethical questions surrounding the welfare of human subjects because of a focus on procedures and documentation; to the contrary, ethical scrutiny is increasing, not decreasing.

Of far greater concern, however, is the contention that IRBs are overstepping their bounds (mission creep) by taking into account issues such as research design and conflicts of interest. Those are precisely the issues that they should examine for human subjects' protection. I have seen experimental designs in IRB proposals that are so flawed and poorly conceived that even if the agent under study worked exactly as hypothesized, the clinical trial would not reveal it. No human subjects should be recruited to participate in such a trial.

Conflicts of interest are of vital concern to IRBs. One only needs to read the recent *Wall Street Journal* revelations about atrial fibrillation ablative studies in which some of the clinical researchers failed to reveal to either the IRB or the patients, through informed consent, that they had a clear financial conflict of interest. This type of "omission" potentially places human subjects in jeopardy and raises the issue of egregious research misconduct.

**DAVID L. FELTEN**

Vice President, Research and Medical Director, Beaumont Research Institute, William Beaumont Hospitals, Royal Oak, MI 48073, USA.

THE EDITORIAL ON "MISSION CREEP IN THE IRB world" (C. K. Gunsalus *et al.*, 9 June, p. 1441) struck a raw nerve. As a scientist approaching retirement after 32 years of research, director of a small nonprofit research institution, and member of two IRBs in the past decade, I now advise students to think twice about getting involved in human research.

I do a great deal of multi-institutional research. It is nearly impossible to deal with a dozen IRBs that review the same protocols when each responds in contradictory ways. Two years ago, one IRB insisted that we could not do what we proposed, and the other IRB involved insisted that we had to do it or they would not approve it. The funded study died. Ten years ago, IRB issues consumed 3 to 5% of my time. Now they consume about 30%.

There is, to my knowledge, not a shred of evidence that the ballooning bureaucracy of IRBs has reduced the number of adverse events or saved a single life. I share the authors' concern that the focus on minor details has diverted discussions from substantive to trivial. It is also diverting scarce funding from research into indirect costs and discouraging talented young scientists from doing human research.

**THOMAS M. VOGT**

Center for Health Research, Hawaii, Kaiser Permanente, 501 Alakawa Street, Honolulu, HI 96817, USA.

### Letters to the Editor

Letters (~300 words) discuss material published in *Science* in the previous 6 months or issues of general interest. They can be submitted through the Web (www.submit2science.org) or by regular mail (1200 New York Ave., NW, Washington, DC 20005, USA). Letters are not acknowledged upon receipt, nor are authors generally consulted before publication. Whether published in full or in part, letters are subject to editing for clarity and space.

### Response

WE ARE PLEASED THAT THE RESPONSES TO our Editorial about IRB mission creep share a strong commitment to protecting human subjects of research. Vogt articulates issues at the center of our concerns: research delayed, deferred, or never attempted; ever-growing costs; trivialization of review; and diversion of resources (*1*).

Felten questions whether IRBs are focusing on form over substance. There is a grow-

ing body of evidence that IRB review, particularly in multicenter trials, is costly and inconsistent and tends to focus on minor matters with little bearing on participant safety (*2*). For example, Rogers *et al.* report on IRBs demanding changes that are inconsistent with federal regulation (*3*). There is also ample, and growing, evidence that some IRBs are going astray and that the costs of review are swelling: Sugarman and colleagues have estimated that IRB operating costs range from $170,000 to almost $5 million annually per institution, depending on the volume of research reviewed. They found a median cost of $740,000, although it is thought that these costs are generally underestimated (*4*, *5*).

This increase in costs, however, is often unrelated to better or more consistent protection for subjects. For example, Green *et al.* document that the costs of securing IRB approval from 43 sites for a 2.5-year multisite observational study totaled 24% of one year's budget and 13% of the total budget. However, "One site exempted it from review (although it did not qualify for exemption), 10 granted expedited review, 31 required full review, and one rejected it as being too risky to be permitted… Twelve sites requested, and two insisted upon, provisions that directly increased the risk to participants" [(*6*), p. 214]. Similarly, Humphreys *et al.* document that 16.8% of the total costs of an eight-site observational trial were devoted to IRB interactions (*7*) but observed that there was no visible effect on human subject protection. The essential procedures of the study never changed substantially, despite exchanges of over 15,000 pages of material among the nine sites."

Finally, we are not against assessment of conflicts of interest, but we believe that there are bodies already constituted at most universities and medical centers better suited to this work. Letting these groups do their job will reduce diversion of IRBs from their core ethical mission.

It is time for all those concerned to find a way to join forces and seek improvements in our ethical systems. We are actively seeking a forum for a consensus conference. Responsible researchers everywhere should be attending to the conduct of IRBs and doing everything possible to buttress their ethical review and minimize their busywork.

**C. K. GUNSALUS, EDWARD M. BRUNER, NICHOLAS C. BURBULES, LEON DASH, MATTHEW FINKIN, JOSEPH P. GOLDBERG, WILLIAM T. GREENOUGH, GREGORY A. MILLER, MICHAEL G. PRATT (MEMBERS OF THE CENTER FOR ADVANCED STUDY ILLINOIS IRB STUDY GROUP)**

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## CORRECTIONS AND CLARIFICATIONS

**News Focus:** "A 'landscape' too far?" by T. Siegfried (11 Aug., p. 750). On page 751, the story stated that physicists' calculations overestimate the vacuum energy by between $10^{60}$ and $10^{120}$ orders of magnitude. The correct figures are between 60 and 120 orders of magnitude. The photo caption on page 751 misidentified Burton Richter as a theoretical physicist. He is an experimental physicist.

**Reports:** "Crystal structure of a divalent metal ion transporter CorA at 2.9 angstrom resolution" by S. Eshaghi *et al.* (21 July, p. 354). On page 357, in the acknowledgments (reference 29), the PDB accession code was omitted: The structural data have been deposited in the Protein Data Bank with accession code 2iub.

**Research Articles:** "Crystal structure of the low-pH form of the vesicular stomatitis virus glycoprotein G" by S. Roche *et al.* (14 July, p. 187). The Protein Data Bank accession number, 2cmz, for the glycoprotein structure described was omitted from the acknowledgments (reference 39).

## TECHNICAL COMMENT ABSTRACTS

### COMMENT ON "Transitions to Asexuality Result in Excess Amino Acid Substitutions"

**Roger Butlin**

Paland and Lynch (Reports, 17 February 2006, p. 990) showed that in *Daphnia pulex*, the ratio of amino acid replacement to silent substitution in the mitochondrial genes is higher in asexual lineages than in sexual lineages. If base-composition bias is maintained by selection, it too should alter following transitions in reproductive mode. Analysis reveals no such change in the genomes of *D. pulex*.

Full text at www.sciencemag.org/cgi/content/full/313/5792/1389b

### RESPONSE TO COMMENT ON "Transitions to Asexuality Result in Excess Amino Acid Substitutions"

**Susanne Paland and Michael Lynch**

Asexual populations experience a reduction in the efficiency of selection when compared with sexual populations. Because asexual lineages of *Daphnia pulex* exhibit no consistent change in mitochondrial base-composition bias, Butlin suggests that this bias is not maintained by selection. On the basis of frequencies of polymorphic directional base changes, we suggest that it predominantly reflects mutation bias.

Full text at www.sciencemag.org/cgi/content/full/313/5792/1389c

PSYCHOLOGY

# How Do Scientists Think?

**David Lagnado**

Have you ever wondered what goes on inside scientists' heads when they formulate a grand theory? Or when they decide what hypothesis to test? How does this differ from the mundane reasoning involved when you explain why your car won't start or choose a birthday present for a relative? More generally, do scientists use the same cognitive mechanisms available to us all (supplemented with formal, conceptual, and technological tools)? Or does scientific thinking require more specialized cognitive abilities, available to only a talented few?

**The Psychology of Science and the Origins of the Scientific Mind**

***by Gregory J. Feist***

Yale University Press, New Haven, CT, 2006. 336 pp. $38, £25. ISBN 0-300-11074-X.

If you are interested in such questions, then Gregory Feist's *The Psychology of Science and the Origins of the Scientific Mind* is the book to read. As the title suggests, Feist (a psychologist at the University of California, Davis) argues the case for a new discipline of "psychology of science" and explores the evolutionary and historical roots of scientific thinking. The first half of the book gives a brief history of three dominant areas in which science itself has been the object of study (history, philosophy, and sociology of science) and reviews a wealth of research implicitly engaged in the psychology of science. This research is divided along traditional lines (biological, developmental, cognitive, personality, and social psychologies), and Feist makes a convincing case for their inclusion in the new discipline. However, his survey lacks an overarching framework and reads more as an assortment from subordinate disciplines. (The desired unification is not helped by the traditional divisions already in place.) If we are envisioning a new discipline, now is a great time to rethink the classic taxonomy—if not to replace it, at least to give it a sound and logical explanation.

What of the origins and precursors of scientific thought? How did we move from preliterate hunter-gatherers who eat their meat raw to sophisticated reasoners with a taste for relativity theory and fine cuisine? In the second half of the book, Feist charts this progression with originality and insight. His speculations on the origins of scientific thinking are particularly impressive and draw well on recent cognitive psychology. He identifies several core components of thought—observation, categorization, pattern recognition, hypothesis testing, and causal thinking—and argues that these were progressively augmented as scientific thinking passed from the preverbal stage through to the explicit research we have today. Critical developments along the way included explanatory thinking (greatly aided by the advent of language), measurement, mathematics, and finally the hallmark of modern science, the experimental method.

This account is well argued and innovative, but more could be made of the dynamic interplay between the key components. For example, both observation and categorization are hypothesis-driven (*1*) and can be influenced by prior causal thinking (*2*). This implies that these components co-develop rather than arise in an incremental fashion. Further support for such co-development is provided by the recent emphasis in cognitive neuroscience on action-based representations (*3*). Thus it appears that our internal models of the world are heavily shaped by the demands of effective action. Indeed, "motor cognition" could be added as a key component in the preverbal stage of scientific thought.

Notably absent from the book are any discussions of the formal or normative models that scientists (or everyday reasoners) ought to use and how these models relate to descriptive models of scientific reasoning. Although it is common to distinguish how people actually reason (descriptive) from how an ideally rational person would reason (normative), both play crucial roles in current psychological research. Normative models serve both as standards against which to appraise human performance and as a framework for understanding cognition (*4*, *5*). For example, there is a growing movement in cognitive psychology and neuroscience that advances a Bayesian perspective on the mind (*6*).

Indeed, one of the appeals of causal maps (which are discussed by Feist in his chapter on cognitive psychology) is that they are formally well defined and normative (*7*). The question of whether people use fully fledged causal graphs (and Bayesian methods), or instead use simplifying heuristics that approximate these norms, is contentious. But there is little doubt that formal models are critical to the development of cognitive models. Moreover, the psychology of science has a special stake in these issues, because the status of normative models is itself keenly debated in current philosophy of science.

Another topic of concern is Feist's attempt to prescribe guidelines for recognizing scientific talent (and its consequences for education and selection policies). He makes much of correlational studies that allow predictions of scientific achievement from intelligence and personality tests and demographics. Such an emphasis is worrying for two reasons: First, there are well-known problems with using correlational studies as a basis for policy interventions. Correlation does not imply causation, and these studies may include all kinds of confounding factors. Second, even if the predictors are valid precursors for the prototypical scientist, would we really want to risk excluding less stereotypical thinkers? Einstein would have fared pretty poorly in terms of early college achievements.

Lastly, there is a hint of paradox in introducing a new discipline to bridge the gap between related disciplines. Once the new discipline is established (complete with specialized conferences and journals), it runs the risk of reducing rather than increasing cross-disciplinary talk. There are now three independent groups that need to share information rather than two, so new bridges must be built, and so on. In the case of the psychology of science, this is not just a theoretical worry. The subdisciplines of psychology already suffer a lack of integration and cross-fertilization; adding another discipline (however much its content spans the divide) might simply add to the problem.

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The barbarism of specialization looms anew.

In spite of these worries, *The Psychology of Science and the Origins of the Scientific Mind* succeeds on many levels. Feist pulls together a vast range of psychological research with clarity and insight, and he advances an intriguing framework for the cognitive origins of scientific thinking. The book makes a strong case for an integrated study of the psychology of science.

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PHYSICS

# The Universe, Too Quickly Toured

**Sean M. Carroll**

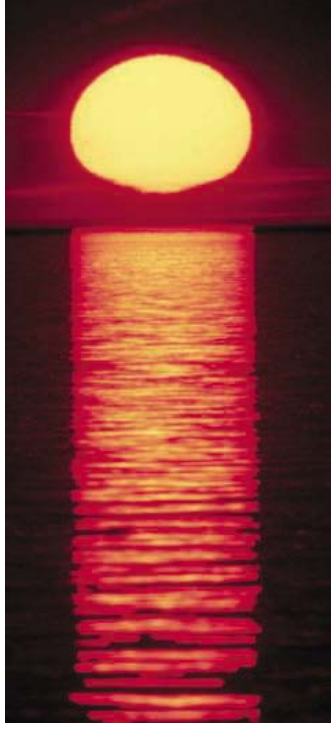
There's no reason why everyone shouldn't understand the basics of quantum mechanics and relativity. These two cornerstones of 20th-century physics have become a basis for our deepest understanding of reality, as well as of great practical importance to familiar technologies from lasers to the global positioning system. And, despite their reputations for being somewhat abstruse and inaccessible, the basic points of each theory can be stated simply enough that an interested person with no technical background in physics should be able to understand them. At a time when science seems both more central than ever and more removed from our everyday world, it is certainly worth the effort to share what we've learned about the workings of nature with interested nonscientists.

We should therefore welcome books like Marcus Chown's *The Quantum Zoo: A Tourist's Guide to the Neverending Universe*. Chown's work is an admirable attempt to delve into the mysteries of these two great theories, quantum mechanics and relativity, and express them in terms that an intellectually curious nonexpert can understand. And for the most part the book succeeds. Chown (a science writer who trained as a physicist) has a pleasant writing style and a facility with simple metaphors and analogies that helps bring difficult concepts into sharp focus.

**The Quantum Zoo**
A Tourist's Guide to the Neverending Universe
***Marcus Chown***
Joseph Henry Press (National Academies Press), Washington, DC, 2006. 212 pp. $24.95, C$27.95. ISBN 0-309-09622-7.

The book is divided into two sections: "Small Things" and "Big Things." In the former, as you might guess, he covers the quantum world, explaining the crucial ideas of superposition and interference, and braving difficult topics such as the uncertainty principle, entanglement, and the collapse of the wave function. Chown moves easily from historical examples such as Young's double-slit experiment and Rutherford's scattering to modern issues such as quantum computers and teleportation. In the second section, devoted to relativity, he swiftly covers the basics of spacetime and relativity, gravitation, and cosmology. The appropriate hot topics are mentioned, if briefly: black holes, string theory, inflation, and dark energy. The brevity of the text is not a shortcoming; not every popular book needs to be a massive and comprehensive tome. The popular audience at which *The Quantum Zoo* is aimed should learn a lot from reading the book and enjoy themselves in the process.

And yet, there is a sense in which the book is a disappointment. There are other books out there, after all, that deal with the topics of quantum mechanics and relativity. To stand out from the crowd, any new entry should have something distinct to offer. It might be the unique insight of a true master of the field, as we find when Richard Feynman writes about quantum electrodynamics or George Gamow writes about the Big Bang. Or it might be an in-depth examination of new and exciting developments in a particular discipline. Or, for that matter, it might just involve bringing a storyteller's eye and a gift for narrative to illuminate a forbidding complex of ideas.



**Tunneling site.** Proton tunelling allows hydrogen fusion in the Sun to occur "even at the ultralow temperature of 15 million degrees."

Unfortunately, *The Quantum Zoo* isn't really distinguished in any of those ways. Chown is a fine explainer, but he doesn't take us over any ground that others haven't trod before. For example, after a good explanation of bosons and fermions takes us up to the connection between spin and statistics, Chown simply admits that this "brings us to the end of what can easily be conveyed without opaque mathematics." Later, after foreshadowing about how superfluid helium can do strange things like crawl up the sides of a container, the book never actually explains why that happens. Dark energy is not explained any more deeply than "the repulsive force of empty space." After whetting our appetites for more substantive explanations, we are left feeling a little unsatisfied.

The primary shortcoming of the book seems to be the lack of some specific point to the project. The subtitle, *A Tourist's Guide to the Neverending Universe*, gives an indication of the unfocused nature of the text. I suspect that Chown could have written an interesting and useful book about quantum mechanics, starting with the basics and going into some detail about modern developments in atomic and molecular physics, quantum information theory, and quantum computation. Or, alternatively, an interesting and useful book about relativity, concentrating on some specific aspect such as gravitational waves, black holes, or dark energy. Instead, Chown's book is competent but uninspiring, a somewhat superficial look at the foundational theories of modern physics. The explanations are clear, and the interested reader will be able to learn quite a lot. But there is not quite any reason to choose *The Quantum Zoo* from among the other titles on the popular-science shelf.

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EPIDEMIOLOGY

# Infectious Diseases: Preparing for the Future

A recent Foresight project report analyzes technological and policy priorities for meeting future challenges of infectious diseases affecting humans, plants, and animals.

D. A. King,[1] C. Peckham,[2] J. K. Waage,[3] J. Brownlie,[4] M. E. J. Woolhouse[5]*

Infectious diseases account for a quarter of all human mortality and a similar fraction of morbidity (*1*). Infectious diseases of crops and livestock cost the global economy uncounted billions of euros every year. On top of this, sudden epidemics of infectious diseases can deliver humanitarian and economic shocks on a scale difficult to absorb. According to the World Bank, the 2003 severe acute respiratory syndrome (SARS) epidemic, which killed fewer then 1000 people, was responsible for an estimated 2% fall in gross domestic product (GDP) across East Asia, and an influenza pandemic could kill millions of people and cost €700 billion (U.S. $900 billion) globally in a single year (*2*). In recent years, there have been numerous outbreaks of livestock and crop diseases costing individual countries billions of euros, for example, foot-and-mouth disease (FMD) in Taiwan and the United Kingdom; bovine spongiform encephalopathy (BSE) in the United Kingdom; classical swine fever (CSF) in the Netherlands; soybean rust in Brazil; Southern corn leaf blight in the United States; and, most recently, avian influenza in Egypt. The United Nations Millennium Development Goals, as well as having explicit targets for reducing the burden of human diseases (particularly HIV/AIDS, tuberculosis, and malaria), also have targets for reducing poverty and hunger, but these are compromised by crop and livestock diseases. In most developing regions, where the impacts of infectious disease are greatest, there is now little hope of meeting any of the Millennium Development Goals by 2015 (*3*).

Governments and international agencies need a vision of how threats such as infectious diseases are likely to evolve in the future so that they can identify effective science and technology strategies to help meet the challenge. Foresight programs, largely originating in Japan and the USA, were put in place precisely to do this. The U.K.'s Foresight program established a series of cross-disciplinary projects to study selected topics in depth, incorporating two key principles (*4*). First, the work has to be based on peer-reviewed science presented in a way that is accessible to nonscientists, and second, decision-makers and government must be engaged from the outset in setting the direction and broad approach of each project.

The latest Foresight project to report (*5*) assessed the projected risks from infectious diseases of humans, animals, and plants over 10- and 25-year horizons. The project focused specifically on detection, identification, and monitoring of disease, aspects widely perceived as neglected and where the development and deployment of new technologies and systems could have major impacts. Earlier disease detection would buy time to allocate resources and, by contrast to current reactive approaches, enable proactive disease management.

The project compared three geographical regions: the United Kingdom (as an example of a developed country), China (a rapidly emerging economy), and sub-Saharan Africa (a developing region). In total, over 300 experts in some 30 countries were consulted by a variety of methods, including Delphi studies (which use formal methods to generate forecasts from groups of experts), expert reviews, workshops, mathematical modeling, and commissioned research.

Eight categories of infectious diseases of the future were identified for which improved detection systems would make a difference over the next 10 to 25 years.

(i) New diseases, such as SARS and BSE, and novel variants, such as H5N1 subtype influenza A, are anticipated to continue emerging. (ii) Infections are becoming resistant to treatment, including antibiotic-resistant bacterial infections, such as tuberculosis and methicillin-resistant *Staphylococcus aureus* (MRSA). (iii) Zoonoses, i.e., infections transferring to humans from animals, are associated with livestock, pets, and, in many cases, with wildlife, e.g., SARS, avian influenza, plague, Lyme disease, and anthrax. This category includes food-borne infections such as *Escherichia coli* O157 or *Salmonella*. Other categories are (iv) HIV/AIDS, tuberculosis, and malaria, the "Big Three" tropical diseases covered by U.N. Millennium Development Goal 6; (v) epidemic plant diseases, such as cassava mosaic virus and banana blight, currently of concern in East Africa; (vi) acute respiratory infections, a category that covers pandemic influenza and a variety of other viral and bacterial infections; (vii) sexually transmitted infections (STIs), including but not limited to HIV/AIDS, which are increasing in incidence in many parts of the world; and (viii) animal diseases, such as FMD, CSF, and Newcastle disease, which remain among the most important barriers to international trade in livestock and livestock products.

Records of mobile phone location could be useful for contact tracing during infectious disease outbreaks.

CREDIT: PHOTOS.COM

The categories are not mutually exclusive and are not intended to be exhaustive, but the list does capture the priority concerns identified by the project. These differed for different regions, e.g., African experts were less immediately worried about new, emerging diseases, and Chinese experts highlighted health care–associated infections as an increasing problem. Overall, it is clear that the infectious disease threat is diverse and dynamic, including "out-of-the-blue" events akin to the emergence of BSE in the United Kingdom in the

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1980s, which implies a need for detection systems that are flexible and adaptable in the face of change.

Among the most important factors expected to influence future changes in infectious disease risks were travel, migration, and trade, which promote the spread of infections into new populations. Particularly worrisome is the trade in exotic species, whether as livestock, pets, crops, garden plants, or food. Changes in land use and agricultural practices, as well as increasing urbanization, were all expected to contribute to changes in infectious disease patterns. Although climate change was a concern, modeling studies suggested that its effect on infectious diseases will be relatively minor over the next 10 to 25 years, becoming potentially more serious in the longer term, both directly, e.g., through changing the distribution of disease vectors and indirectly, e.g., by changing patterns of land use and agriculture. Poor governance and the loss of capacity to recognize and respond to infectious disease problems is a major issue for sub-Saharan Africa in particular. The report also anticipated that, despite the alarming spread of antibiotic resistance, widespread overuse and misuse of antibiotics and other drugs would continue, further exacerbating drug resistance problems. Taken together, these drivers of changing infectious disease risk have complex and interrelated effects, making the prediction of future risk extremely uncertain and again underlining the need for flexible detection systems.

A wide range of technological advances, from remote sensing to nanotechnology, were reviewed and ultimately four were selected for detailed consideration (*5*). Novel information technologies for the capture, analysis, and modeling of data are already being developed. These will allow data to be collected electronically from hand-held devices or remote sensors and analyzed and modeled in real time. Genomics and postgenomics approaches will allow the rapid characterization of pathogens. Mass screening of people, animals, and plants in transit should become feasible through noninvasive detection systems for volatile organic chemicals or atypical electromagnetic profiles. Portable devices will become available for diagnosing infections in individual patients, animals, or plants, satisfying a growing demand for cheap, quick, easy-to-use, over-the-counter products, perhaps resembling today's home pregnancy test kits. Some of these technologies will be generic, such as "lab-on-a-chip" screening for a range of infectious agents, nonspecific diagnostics based on detecting activated immune responses, or simple tests to differentiate between viral and bacterial infections to aid the appropriate prescription of antibiotics.

Better disease detection capability is vital but will present challenges as well as opportunities. New technologies must be embedded within functional national or international surveillance systems. In practice, it is not clear whether or how government agencies would gain access to valuable data obtained through the widespread use of self-administered tests. Other kinds of information, e.g., records from mobile phones or traffic cameras to follow movements, are potentially valuable for disease-control purposes, particularly in outbreak situations, but the public might not accept their use for this purpose. There is always the danger that disease data could be used to discriminate against individuals without providing any benefit or compensation. Similarly, technologies deployed by developed countries could disadvantage developing countries by restricting travel and trade on the basis of the presence or even the suspicion of an infectious disease. Finally, everywhere in the world, better disease data might cause public alarm and raise expectations of effective action, whether or not this is realistic. Surveillance needs to be linked operationally to an appropriate response. For example, the Global Plan to Stop TB relies on the combination of case detection and directly observed therapy (DOTS) (*6*), and plans to combat influenza involve surveillance, drug delivery, and vaccine production (*7*).

The Foresight project highlighted the importance of fostering interdisciplinary approaches to infectious disease research that transcend traditional intellectual boundaries, such as those between medicine and veterinary medicine or among virology, bacteriology, mycology, and parasitology. A better understanding of patterns of infectious disease also needs input from disciplines as diverse as anthropology, economics, and climatology. Quantifying these relations and understanding their dynamics require inputs from statistics and mathematics. Health systems research is needed to understand how new technologies can be used most effectively and must include consideration of the needs, expectations, capabilities, and sensitivities of end users and other stakeholders.

The Foresight project also highlighted a number of key choices for policy-makers. Among the most important of these was that more extensive international coordination should be sought by building on the work of the World Health Organization, the World Organization for Animal Health, the Food and Agriculture Organization, and other agencies. For this to be effective it is essential that data and biological material can be rapidly collected and openly shared. Recent success in combating SARS illustrates what can be achieved (*8*). Currently, the extreme disparity in detection and disease management capabilities between nations, reflecting massive inequalities in wealth, often exacerbated by poor governance, seriously hinders our ability to tackle infectious disease problems quickly.

In the 1940s, the U.S. government created a national rapid response capability, now the Centers for Disease Control and Prevention, in response to malaria originating from Africa. Sixty years later, Africa and other regions still do not have equivalent capabilities of their own, or the infrastructure, skills, and training base to support them. The Foresight report advocates the benefits of a system of regional reference and coordination centers (RRCCs). These would form a network of high-quality laboratories linked to one another, to existing facilities, and to appropriate partners in developed countries. Collectively, the network would have the physical and human resources to support infectious disease detection, identification, and monitoring and the development of suitable technologies and appropriate training programs. The network would aim to cover a range of diseases of humans, animals, and plants, exploiting the commonalities across different disease problems and so being more cost-efficient than traditional "one-disease-at-a-time" initiatives. It is ever more apparent that the benefits of such a facility would extend not just nationally or regionally, but globally; surveillance for infectious diseases has become a collective responsibility and requires a collective investment. The recent statement from the G8 summit in Russia, which calls for "tangible progress" on international disease surveillance (*9*), will, we hope, encourage the international community to make that investment.

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GEOPHYSICS

# Another Nail in the Plume Coffin?

At least one chain of hot-spot volcanoes is not caused by a plume rising up from the core-mantle boundary, calling for a reexamination of the plume hypothesis.

**Marcia K. McNutt**

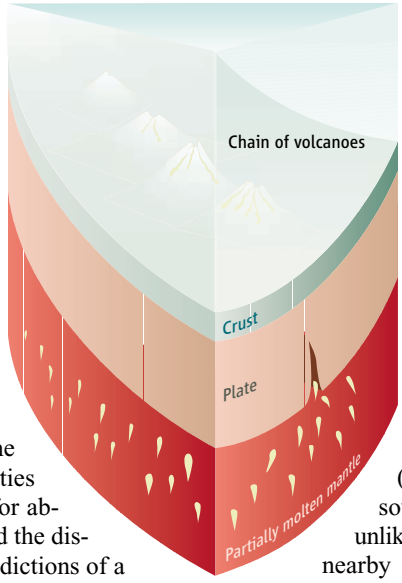
Scientists love beautiful theories—the kind that are elegant, predictive, and have few free parameters. And they hate it when theories like that prove to be wrong. It is thus with much kicking, dragging, and screaming that geoscientists are being brought to the realization that all might not be well with the concept of mantle plumes.

According to the plume hypothesis, linear chains of volcanoes that form at "hot spots" away from the boundary zones of tectonic plates are caused by hot, buoyant jets that detach from the thermal boundary separating Earth's core from its mantle. The latest attack on this hypothesis comes from Hirano *et al.* on page 1426 of this issue (*1*). The authors describe a small chain of hot-spot volcanoes off the Japanese coast that almost assuredly cannot have been formed by narrow, deep-Earth upwellings.

Enhanced online at www.sciencemag.org/cgi/content/full/313/5792/1394

The plume hypothesis was originally proposed by Jason Morgan (*2*), hot on the heels of the plate tectonic revolution. According to the latter theory, most geologic activity—such as volcanic eruptions, earthquakes, and mountain building—is concentrated at the margins of the dozen or so major plates that move together, move apart, or slip past each other on the surface of our planet. Morgan and others noted that island chains such as the Hawaiian Islands and the Canary Islands consist of young volcanoes in the middle of the tectonic plates and therefore cannot be explained by plate tectonics. Moreover, many of these island chains, particularly those on the Pacific plate, show a monotonic increase in age in one direction. Morgan hypothesized that these midplate volcanoes, termed hot spots, are the surface manifestation of mantle plumes that rapidly (to a geologist's way of thinking) rise up from the core-mantle boundary.

Morgan's hypothesis has several attractive features. First, it suggests that the study of hot spots will provide clues to the chemistry and dynamics of Earth's interior, which would otherwise be masked from scrutiny by the rigidity and surface-dominated processes of plate tectonics. Second, the orientation and rate of age progression of hot-spot volcanoes reveal the direction and speed of absolute motion of the tectonic plates, whereas only relative motion could be inferred from the properties of the plates themselves. Scientists therefore immediately started to explore the implications of the plume model for properties of Earth's interior and for absolute plate motions, and the distinction between the predictions of a hypothesis and observed facts quickly became blurred.




**A new kind of volcanism?** In this three-dimensional cartoon, a crack in a flexed plate provides a conduit through which buoyant pockets of partial melt in the upper mantle can reach Earth's surface.

But troubling inconsistencies began to emerge. The absolute motions of plates inferred from hot spots on different plates did not agree, prompting the proposal that the plumes "blow in the mantle wind" (*3*). Rocks from hot-spot volcanoes with different ratios of noble-gas isotopes were thought to tap separate reservoirs in the mantle that were isolated from intermixing. Unfortunately, as the number of isotopic tracers grew, additional reservoirs were required (*4*), taxing the ability of geodynamic models to keep them isolated over geologic time scales. Furthermore, because of their small dimensions, plumes proved difficult to detect in tomographic images of the mantle. Advanced techniques do suggest the upwelling of hot material from the lower to the upper mantle beneath some hot spots (*5*), but not beneath others. Are these problems simply the maturing of a valid theory to deal with the complexity of the real planet, or are they the signs of a paradigm in crisis?

Although insignificant in size on a planetary scale, the small seamounts described by Hirano *et al*. are important because they are young hot-spot volcanoes (1 to 8 million years old) that erupt through a very old plate (135 million years old), away from any plate boundaries. Petrologic data confirm that the volcanoes tap a region of the mantle more than 100 km deep that contains very small percentages of melted material. The position of the volcanoes in the flexural trough seaward of the outer rise of the Japan trench suggests that elastic bending of the plate has opened up conduits through which the partial melt can rise to the surface (see the figure). A plume source for these volcanoes is unlikely in any case, because the nearby subduction of the Pacific plate creates widespread downwelling that would block the rise of a mantle plume.

The notion that the upper mantle beneath the plates might contain small amounts of partial melt just about everywhere, and that all it takes is a crack in the plate for that melt to rise to the surface, has been previously proposed as an alternative to the plume hypothesis (*6*). However, prior examples of hot spots used to illustrate this concept were not as compelling, because there was no obvious mechanism for cracking the plate and they were located away from sites of obvious large-scale downwelling.

Does the plate-cracking mechanism proposed by Hirano *et al.* only explain this one small chain of volcanoes, or can it also explain other chains of hot spots? Hirano *et al.* argue that they have found a new and different kind of hot-spot volcano. They base their argument on the observed ratios of isotopes of inert gases. According to plume theory, plumes tap a deep reservoir rich in rare gases, whereas the mid-ocean ridges obtain melt from a reservoir in the upper mantle that is depleted in rare gases. The volcanoes described by Hirano *et al*. have more depleted isotopic ratios, such as those found in mid-ocean ridge basalts; according to plume aficionados, this property would put them in a category different from hot spots such as those in Iceland and Hawaii. However, other interpretations of the rare gas isotopes would not necessarily require distinct mantle sources (*7*).

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It would be an overgeneralization at this point to conclude that plume theory must be discarded, but the study by Hirano *et al.* proves, beyond a reasonable doubt, that at least one chain of hot-spot volcanoes is not produced by a plume. Therefore, it is time to critically reexamine the widespread interpretation of midplate volcanism in terms of mantle plumes and all the geodynamic, geophysical, and geochemical inferences that have been built on the plume paradigm.

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PSYCHOLOGY

# Is She Conscious?

Brain imaging reveals unexpected activity in a patient clinically diagnosed as being in a vegetative state, raising questions about the properties of consciousness.

**Lionel Naccache**

"Is she understanding my words and feeling my caresses?" is a question constantly asked by relatives and caregivers of comatose and vegetative state patients in intensive care units throughout the world. Probing residual mental function in such critical situations poses major medical and ethical issues. Our current answers to this question are mainly based on detailed behavioral and neurological observations, but this approach may be blind to inner active mental processes. Since the late 1980s, several neurophysiological correlates of cognitive processes have been proposed to better assess the existence of this covert mental life (*1*). This approach has aimed to discriminate among comatose or vegetative state patients those who are still cognitively active. On page 1402 of this issue, Owen *et al*. (*2*) use functional magnetic resonance imaging (fMRI) to infer the psychological processes at work in such patients during mental imagery tasks that do not elicit otherwise observable behavior.

The authors studied a 23-year-old woman who sustained a severe traumatic brain injury in a road traffic accident. After an initial comatose state (defined as an unarousable unresponsiveness state), she opened her eyes and demonstrated sleep-wake cycles. However, even during the waking periods, she was unresponsive (for example, to visual or auditory stimuli) and did not manifest spontaneous intentional behaviors. These signs are diagnostic of a vegetative state.

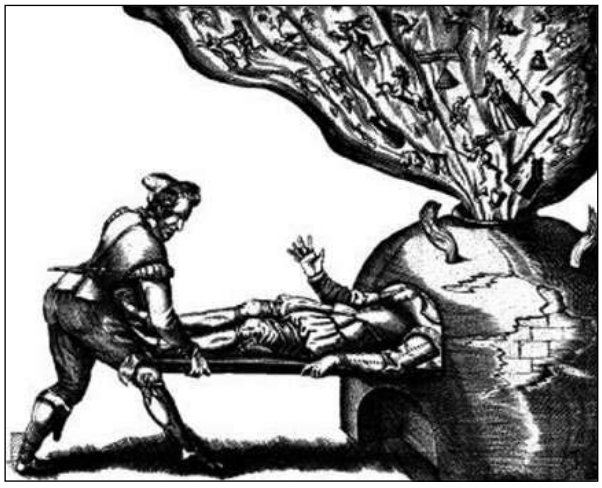
In a first experiment conducted 5 months after the accident, Owen and colleagues presented spoken sentences to the patient while recording neural responses with fMRI. Speech-specific brain regions were clearly activated while the patient listened to these sentences, as compared to acoustically matched noise sequences. Moreover, sentences containing ambiguous homophone words (for example, creak/creek) activated an additional left inferior frontal lobe region known to subserve the selection of semantic knowledge among competing alternatives (*3*). In sharp contrast to her behaviour, which was suggestive of poor cognitive abilities, this patient could process external auditory information involving language.

In a second experiment, the authors engaged the patient in two mental imagery tasks by asking her either to "imagine visiting the rooms in your home" or to "imagine playing tennis." The result of this second fMRI investigation is quite spectacular. Patterns of brain activation observed during the 30-s period of each task were highly suggestive of an active mental performance relevant to the task. In the spatial mental imagery task, Owen *et al*. observed neural activations within a network including the parahippocampal gyrus, posterior parietal cortex, and lateral premotor cortex. When the patient was asked to imagine playing tennis, strong activations were recorded in the supplementary motor areas that control motor responses.

The observed brain activation patterns are the classic neural correlates of these two mental imagery tasks. Indeed, statistical parametric maps of brain activation observed in the patient were indistinguishable from those recorded from a group of conscious control subjects.

Despite the patient's very poor behavioral status, the fMRI findings indicate the existence of a rich mental life, including auditory language processing and the ability to perform mental imagery tasks. On one hand, this single case makes a strong argument for the development of fMRI and other neurophysiological tools (such as monitoring electroencephalogram brain responses to external stimuli) to evaluate cognition in such patients. On the other hand, we should not generalize from this single patient, who suffered relatively few cerebral lesions, to most other vegetative state patients, who typically have massive structural brain lesions.

Is this woman conscious? Most current behavioral and neuroimaging evidence suggests that conscious processing is abolished during a vegetative state. Such patients do not report mental states, nor do they spontaneously engage in intentional actions or interactions with their environment, two key properties of con-



CREDIT: MATHAUS GREUTER

**Imaging imagination.** Shown is part of the engraving, "The Physician Curing Fantasy," by Mathaus Greuter (1564–1638).

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scious processing in humans. Moreover, several studies have reported residual local and specific brain activation patterns in vegetative state patients, whereas long-range neural integration observed during conscious processing was lacking (*4*, *5*). Nevertheless, on the basis of their findings, Owen *et al*. argue that the patient in their study was probably conscious of herself and her surroundings during fMRI testing. This hypothesis opens another issue: If this patient is actually conscious, why wouldn't she be able to engage in intentional motor acts, given that she had not suffered functional or structural lesion of the motor pathways?

The debate over whether vegetative state patients can engage in conscious processing is reminiscent of the Turing test in artificial intelligence: Can we distinguish a conscious human from a computer solely on the basis of a question-answer method (*6*)? Adapting the Turing test to the present debate, we might ask: Can we determine whether a person is conscious solely on the basis of a question–brain activation method? Whereas these questions have stimulated intense philosophical debate about artificial intelligence, most cognitive neuroscientists have adopted a more naturalistic approach. Consciousness is univocally probed in humans through the subject's report of his or her own mental states. A subject who reports, "I read the word consciousness on this page," can be considered as conscious (*7*). The ability to report one's own mental state is the fundamental property of consciousness.

Owen *et al*. did not directly collect such a subjective report. When conscious reporting is not possible, an alternative solution is to examine the three other psychological attributes of conscious processing: (i) active maintenance of mental representations; (ii) strategical processing; and (iii) spontaneous intentional behavior (*8*). Clearly, one of the most impressive aspects of the work by Owen *et al*. is the demonstration that activation of task-related neural networks is actively maintained. During each experimental task, instructions were delivered only once, and the corresponding neural network remained activated throughout the entire 30-s period. In contrast, unconscious mental representations observed in many clinical and experimental contexts are fleeting, lasting a few seconds or less (*9*–*11*).

Though not totally convincing on the issue of consciousness, the Owen *et al*. work paves the way for future functional brain-imaging studies on comatose and vegetative state patients. One can imagine probing each of the psychological properties of conscious processing listed above, and even trying to collect subjective reports by modifying the experimental paradigm.

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10.126/science.1132881

ECOLOGY

# How Does Climate Change Affect Biodiversity?

The most recent and complex bioclimate models excel at describing species' current distributions. Yet, it is unclear which models will best predict how climate change will affect their future distributions.

**Miguel B. Araújo and Carsten Rahbek**

Over the past 100 years, Earth's climate has become warmer and precipitation regimes have changed. Can biologists predict the effects of these changes on the distributions of species?

Conservation strategies for managing biodiversity have traditionally assumed that species distributions change relatively slowly, unless they are directly affected by human activities. However, there is a growing consensus that these strategies must anticipate the impacts of climate change (*1*, *2*). Conservationists must therefore assess both current and future distributions of species. Numerous new bioclimatic models estimate relationships between the distributions of species and climate. However, the decision of which model to use has generally been ad hoc, and there is little consensus regarding the relative performance of these models.

Bioclimatic modeling has been driven by a pragmatic desire to obtain results that are useful for biodiversity management (*3*, *4*). The models are based on some problematic ecological assumptions—for example, that species distribution and assemblages are in a constant steady-state relationship with contemporary climate—that, despite being clearly acknowledged (*5*), remain unresolved. However, there has been even less emphasis on understanding which models best predict species distributions and why.

The proponents and architects of some of the most prominent bioclimatic models recently joined forces to test the predictive uncertainties of their models and to identify the techniques best suited for modeling current species distributions. Elith *et al*. have now published the first results in *Ecography* (*6*). Sixteen models were tested on climate and species distribution data from five continental regions. In contrast to many previous studies, data for testing the models were collected independently.

The models with the best performance were the most recent and complex ones and fell into two groups: machine-learning programs that seek to obtain a stable selection of predictors from a larger range of alternatives, and community models that simultaneously analyze all species in relation to environmental parameters and then calibrate model coefficients for individual species. In contrast, some of the most widely used models for modeling species distributions, such as GARP (which uses a genetic algorithm) and BIOCLIM (which uses an envelope approach), performed poorly under the criteria used to evaluate them.

One critical question is whether models that can successfully predict current species distributions also provide robust predictions of future distributions under climate change. (This question is not addressed by Elith *et al.*,

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who focus on current distributions.) Different bioclimatic models can produce highly variable predictions of species-range shifts (*7*–*11*), and there is a poor correlation between a model's ability to fit present and future distributions (*12*). For example, Pearson *et al.* (*9*) applied nine bioclimatic models to predict the distributions of four South African plants under current and future climates. Predicted distribution changes varied from 92% loss to 322% gain for one species; similar variability was recorded for the other species. In another study, observed and predicted changes in the distributions of British breeding birds differed markedly for 90% of the 116 birds modeled (see the figure) (*8*).

Evaluating model performance under climate change requires a paradigm shift, because there are no data against which predictions of future ranges can be tested (*12*). One way to overcome this problem is to make use of backward predictions, or "hindcasting." Here, models are calibrated with current species-climate relationships and are then tested with reconstructed species distributions from the fossil record. This approach has been used to test whether climatic requirements of species remain stable over time (*13*, *14*). However, hindcasting is only feasible for a few species and regions for which a good fossil record is available.

The predictive ability of models can also be tested through "space-for-time" substitution. Here, bioclimatic models are calibrated with data from one region, and predictions are tested with distributions of species from other regions. Randin *et al.* recently illustrated the principle by predicting plant species distributions in the Austrian Alps based on knowledge of species-climate relationships in the Swiss Alps and vice versa (*15*). They found that predictions from generalized linear models (which impose a theoretical response curve) were more easily transferable in space and time than generalized additive models (which produce data-driven response curves). However, the latter yielded more precise predictions in the regions where the models had been calibrated.

Do data-driven, machine-learning, and community models provide more precise predictions of species distributions in a given region because they overfit the data? Does model precision come at the expense of generality, that is, the ability to predict species distributions in different regions or times? And do theory-driven response curves improve the generality of models? The results of the two studies (*6*, *15*) call for a second generation of studies to test predictions of bioclimatic models under climate change.

Predictions of future distributions of species from bioclimatic models may fail because of uncertain predictions of local climate change, inaccurate estimates of the climatic tolerance of species, and unforeseen evolutionary changes in populations (*16*). We will never be able to predict the future with accuracy, but we need a strategy for using existing knowledge and bioclimatic modeling to improve understanding of the likely effects of future climate on biodiversity.


PHOTO CREDITS: RED-BACKED SHRIKE, HERBERT ZETTL/ZEFA/CORBIS; TURTLE DOVE, ERIC WOODS/OSF/ANIMALS ANIMALS

**A mixed picture of model performance.** Observed and predicted distributions of the red-backed shrike (*Lanius collurio*) and turtle dove (*Streptopelia turtur*) in Britain. Bioclimatic models predict the distributions in the 1970s reasonably well, but fail to predict the contraction of the range of the red-backed shrikes in the 1990s. However, the contraction of the range of the turtle dove is successfully predicted by models. Maps were produced with data and generalized linear models from (*8*).

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CELL BIOLOGY

# Peptides, Scrambled and Stitched

Cells that display peptide antigens on their surfaces for immune recognition usually cut large proteins into smaller pieces, but can also splice these to make larger, scrambled peptides.

**Nilabh Shastri**

How does our immune system know whether a virus is lurking inside a cell, whether a normal cell has turned cancerous, or whether a cell is from an organ that was transplanted from an unrelated donor? Such cells are distinct from our own normal cells, but the differences are often hidden deep inside a cell's genome. However, our immune system has the remarkable ability to survey these differences through a process called antigen presentation. On page 1444 of this issue, Warren *et al*. (*1*) reveal an unexpected mechanism by which cells generate these presented antigenic peptides, increasing the breadth of proteins generated from the genetic code.

Antigenic peptides are normally 8 to 10 amino acids and are exposed on the cell surface (see the figure) (*2*). They are derived from normal self-proteins and from foreign or abnormal ones, such as a viral protein in an infected cell or a mutant protein in a cancer cell. Killer T cells of the immune system have evolved to recognize these peptides through antigen receptors. The peptide thus acts as a flag for killer T cells to recognize, triggering the elimination of an undesirable "foreign" or abnormal cell. Unfortunately, the same mechanism also rejects transplanted organs and can cause autoimmunity as well. In identifying an antigenic peptide associated with a human leukemia that triggers killer T cells, Warren *et al*. have discovered a new twist to how antigenic peptides are generated.

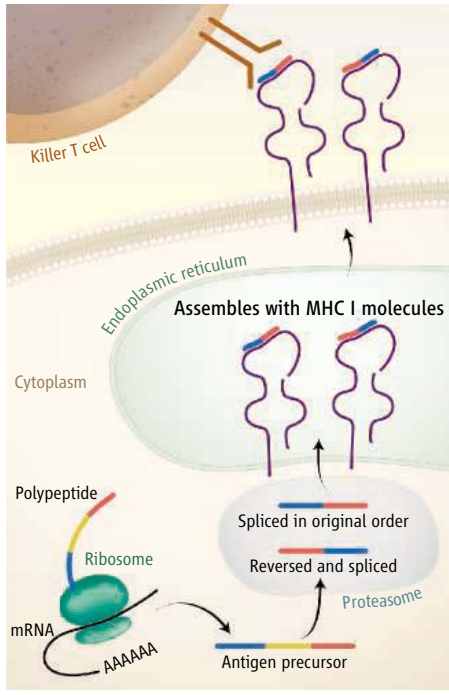
Identifying the particular peptide that is recognized by a killer T cell is challenging. Tens of thousands of peptides are collected in a cell's endoplasmic reticulum and brought to the cell surface by chaperones called major histocompatibility complex class I (MHC I) molecules (pMHC I refers to its peptide-bound form). Only a few copies of a particular pMHC I molecule suffice to engage the T cell receptor on a killer T cell and initiate an immune response (*3*).

The exquisite sensitivity with which killer T cells detect rare pMHC I molecules makes it possible to screen a gene library generated from an antigen-presenting cell. Cells with the appropriate MHC I but lacking the antigen are given small pools of genes from such a library and are then tested for an acquired ability to activate killer T cells. This screen eventually yields a single gene whose sequence reveals the predicted protein. Identifying the actual peptide usually involves testing fragments of the isolated gene and/or candidate synthetic peptides for their ability to activate killer T cells.

Since the late 1980s, this expression cloning method has successfully identified numerous genes and their pMHC I products that elicit killer T cell responses. For most gene products, the final peptide is a contiguous sequence of amino acids within the predicted protein sequence. That the peptide should be contiguous is also consistent with our understanding of the cellular antigen presentation pathway because these peptides are made by cutting the polypeptide precursor to create the exact carboxyl- and amino-terminal ends of the final peptide (*2*).

Running against this history and current concepts of how the peptides are generated, Warren *et al*. now identify an antigenic peptide that is not encoded by a contiguous stretch of codons within its corresponding gene. Rather, they found that the final peptide is stitched or spliced together from a tetra- and a hexapeptide. The evidence indicates that the peptide is spliced within the cell's multicatalytic proteasome, which is paradoxically famous for its appetite to destroy proteins. The major surprise is that unlike earlier discoveries of spliced peptides (*4*, *5*), the final decapeptide is a scrambled result of putting together two smaller peptides in reverse order!

The discovery of scrambled and stitched peptides adds another source for expanding the peptide repertoire displayed by MHC I molecules. Other unexpected sources include peptides derived from cryptic translation of noncoding regions of the genome or alternative translational reading frames (*6*). However, the idea of stitching together different bits of information to maximize diversity is not new to the immune system and is in fact essential for dealing with an even more diverse and constantly changing microbial universe. Thus, the vast repertoires of antibodies and T cell antigen receptors are constructed by stitching together different DNA segments. Likewise, differential splicing of primary RNA transcripts converts membrane-bound antibody molecules present on B lymphocytes into those that are secreted. The DNA rearrangement mechanisms are used in all known immune systems from the earliest jawed fish to humans, and RNA splicing occurs in other eukaryotic cells as well.




**The antigen presentation pathway.** Antigenic polypeptide precursors move to the proteasome where they are cut. Smaller peptides are spliced in either the same or reverse order. These peptides are then transported into the endoplasmic reticulum, where they are trimmed, assembled with MHC I molecules, and then expressed on the cell surface. Killer T cells recognize specific pMHC I on the cell surface and eventually cause lysis of the pMHC I–presenting cell.

CREDIT: P. HUEY/SCIENCE

By contrast, examples of protein splicing are rare. The best known are the inteins in lower organisms such as fungi (*7*). Inteins are parasitic genetic elements that insert into the host's genome and can be expressed as polypeptides. Because inteins also contain an autocatalytic activity, different fragments of the intein polypeptide can be spliced together. However, unlike inteins, which do not need extraneous help, spliced peptides require the catalytic activity of the proteasome (*1*, *4*, *5*). It is puzzling that the proteasome, which has been described as the

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"chamber of doom" for its ruthless chopping of proteins, is now also implicated in the creative activity of joining shorter peptides into a longer peptide (*8*). A possible explanation for this function might be that, like all enzymes that catalyze reactions in one direction, it can—in principle and under some circumstances—catalyze the same reaction backward. Alternatively, specific sequence or structural features of the final peptide itself, or its flanking and/or intervening amino acids, might favor the splicing reaction. Further experiments are needed to resolve how proteasomal splicing works.

Irrespective of the still obscure rules for making noncontiguous peptides, their existence shows that genetic information can be scrambled and yet be useful. Cells do not simply discard scrambled peptides, and, like defective or cryptic translation products, use them to add to the pMHC I repertoire (*2*, *9*). This view implies that such scramblings are not random accidents and would have to be generated in other cell types as well. In particular, cells constituting immune organs that are responsible for eliminating self-reactive T cells must generate the same scrambled peptides. This is to ensure that autoimmunity will not result from the sudden appearance of previously unseen pMHC I in some tissues. What fraction of the total pMHC I repertoire represents cryptic translation products or spliced and scrambled peptides, and whether and how they might be regulated, remain important unanswered questions. Nonetheless, it is clear that peptides from these sources are a functional aspect of the antigen presentation mechanism that keeps an eye on the genome, including its unexpected and unpredictable products.


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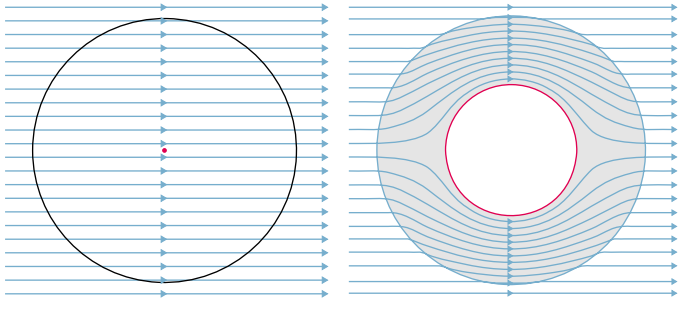
APPLIED PHYSICS

# Waves on the Horizon

Emerging materials can influence the dissipation, dispersion, and refraction of light, producing resonant effects that allow intricate control of electromagnetic waves for new applications.

**Ping Sheng**

The ability to localize waves and create artificial structures capable of manipulating waves are arguably two of the most important developments in optical physics during the past few decades. These advances have been made possible by the fabrication of photonic and phononic crystals (periodic structures that modify the characteristics of photons and phonons, creating frequency band gaps in which no waves can propagate), as well as metamaterials (in which wave properties beyond those intrinsic to the underlying component material properties can be structurally induced). A recent conference of the Optical Society of America (*1*) brought together researchers working in these two areas and highlighted the latest advances. If Lord Rayleigh, who laid the groundwork for much of wave physics a century ago, were alive and had attended the meeting, he might even be surprised to find that what were formerly regarded as "constraints" in wave studies have been knocked down, uncovering new territory in science and technology. In particular, the index of refraction can now take both positive and negative values, waves can be manipulated in ways not thought possible before, and multiple scattering can cause a wave to alter its basic character while retaining phase memory.



**Hidden realms.** (**Left**) Blue lines indicate the flow of electromagnetic energy through a region of space. (**Right**) The shaded region has been subject to a coordinate transformation. The center of the region denoted by the red dot at left is transformed to the red circle. The region inside the red circle is cloaked and cut off electromagnetically from the region outside. Outside the shaded area, the flows are identical in both panels, implying electromagnetic nondetectability of objects inside the cloaked region.

Maxwell's equations contain two material parameters, the dielectric constant $\varepsilon$ and magnetic permeability $\mu$. If the material is anisotropic, $\varepsilon$ and $\mu$ are $3 \times 3$ matrices in general. Under a coordinate transformation, the form of Maxwell equations should remain invariant, but $\varepsilon$ and $\mu$ would carry the information regarding the original values plus the relevant coordinate transformations. If all the values and anisotropy characteristics of $\varepsilon$ and $\mu$ are physically realizable, as implied by the advent of metamaterials, then theoretically one can access the many possibilities afforded by the coordinate transformations. Leonhardt (*2*) and Pendry *et al.* (*3*) have pointed out one such possibility that might have come from *Star Trek*—electromagnetic cloaking. The idea is to create a "hole" in the transformed coordinate system in which an object (or objects) can be hidden from detection (see the figure). The hole is not an electromagnetic vacuum but rather a complete separation of electromagnetic domains into a cloaked region and those outside. A cloaked object thus cannot communicate with the outside, and vice versa. A different form of cloaking has also been proposed by Milton and Nicorovici (*4*). By considering a polarizable line dipole in the vicinity of a coated cylinder with core dielectric constant $\varepsilon_c$ and surface coating dielectric constant $\varepsilon_s = -\varepsilon_c$, they have shown that under the action of an external quasistatic transverse magnetic field, both the coated cylinder and the polarizable line dipole are invisible—that is, in the cloaking region, the polarizable line dipole produces no induced dipole moment. The reason for this strange behavior is that any induced moment would be

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canceled by the resonant response of the coated cylinder. Otherwise, the energy can diverge (*4*, *5*). Because a single polarizable line dipole produces no induced moment, it follows that a collection of such objects would also produce no moment, and hence be invisible.

To realize all these possibilities requires ingenious materials design supported by advances in technology. There is a plethora of reported advances in both wave-functional materials fabrication, as well as the realization of related phenomena. Because many of the wave-functional effects are associated with resonances, overcoming the limitations imposed by dissipation and dispersion effects (meaning that the desired phenomena are realizable only within a narrow frequency window) represents the most urgent challenge. In this respect, the successful achievement of a photonic crystal optical cavity $Q$ value on the order of $10^6$ by Noda's group in Kyoto University (*6*) is noteworthy for foreshadowing the potential applications. There are also efforts to realize a negative refraction index through structural means, such as extreme anisotropy (*7*) and chiral materials (*8*), in addition to photonic crystals. An interesting proposal is to compensate the resonance-induced dissipation with an optical gain medium (*9*), which can be pumped separately. The degree to which these efforts are successful would set the scenario of future wave technology.

Wave localization in the Anderson sense (that is, localization of waves as they scatter in a random medium) is generally characterized spatially by an exponentially decaying wave function. However, if one uses a pulse to probe a medium with localized states, then it can be shown theoretically that there are also characteristic time-domain signatures (*10*). Recent experiments by Storzer *et al.* (*11*) have shown that by measuring time-resolved photon transport through $TiO_2$ powder samples, one can detect clear deviation from the diffusive behavior that is expected from multiple scatterings (*12*). Moreover, it was reported during the meeting that the deviation can be explained by a time-dependent diffusion constant $D$ that approaches a $\sim 1/t$ behavior. If $r^2 \sim Dt$, then heuristically $D \sim 1/t$ implies a saturation length—the localization length. The photon mobility edge, the optical analog to the electronic metal-insulator transition, may be within reach.

Random systems are usually characterized by probability functions. Thus, wave localization, a manifestation of multiple scattering of waves in random media, has been mostly studied by focusing on the mean behavior, just as diffusion is the mean behavior of a random walker. A shift away from this focus is represented by the study of the "connectivity" of localized wave functions in a single (finite) random configuration. In a one-dimensional layered system, a connected state consisting of multiple localized wave functions (*13*) with roughly the same energy and equally spaced across the sample is denoted a "necklace." Such necklaces would carry most of the wave flux through the sample, because they represent short circuits in an otherwise insulating sample. In two separate experiments, one in the microwave regime by Sebbah *et al.* (*14*), and one in the optical regime by Bertolotti *et al.* (*15*), these necklace states in a one-dimensional layered system were observed. The true significance of these states may lie in the three-dimensional mobility edge, where in analogy with percolation the connected localized states would play a role similar to that of the percolating backbone, which has density measure zero (because of its fractal geometry) but nevertheless carries all the flux.

As the title of the meeting "From Random to Periodic" implies, some convergence of the two developments may be inevitable, or even anticipated. Challenges remain, however, in identifying the nontrivial intersections, from which new physics and phenomena may emerge.


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PHYSICS

# Entangled Solid-State Circuits

**Irfan Siddiqi and John Clarke**

Quantum tomography is used to determine the entangled state of two coupled superconducting qubits, a step forward for solid-state quantum computing.

A fundamental tenet of quantum mechanics is the idea that two spatially separated objects exhibit correlations in observable physical properties that cannot be explained by any classical theory. Troubling even Einstein, this "spooky action at a distance" (*1*)— known as entanglement—is fundamental to quantum information science and directly related to the enhanced computing power of a processor based on quantum bits (qubits). What is remarkable is that solid-state electrical circuits containing as many as $10^{11}$ atoms can be engineered to exhibit quantum behavior and are well described by the quantum formalism originally developed for individual atoms and photons. One can construct such qubits from thin films using conventional semiconductor fabrication techniques, making them attractive for eventually realizing a quantum computer with many qubits.

With these solid-state "atoms on a chip," one can prepare arbitrary superpositions of single-qubit states and manipulate them with microwave radiation to observe clear signatures of quantum coherence familiar in atomic physics and nuclear magnetic resonance (*2*–*5*). Coupling two or more qubits together results in entangled states with energy spectra that exhibit features such as avoided crossings (*6*) predicted by quantum mechanics. Verifying that two qubits are unambiguously entangled is, however, a delicate task and requires sophisticated benchmarks such as quantum state tomography (*7*). This method involves a series of measurements (analogous

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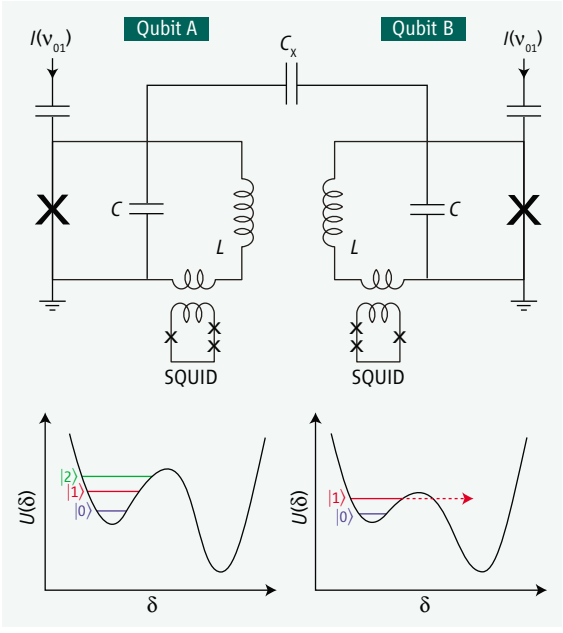


**Coupled qubits.** (**Top**) Simplified schematic of two phase qubits coupled by a capacitance and driven by microwave current sources $I(\nu_{01})$ (**Bottom left**) Potential-energy wells of a fictitious particle versus phase difference δ. The barrier height and level spacing are adjusted by an external magnetic flux. Microwaves induce transitions between the states $|0\rangle$ and $|1\rangle$ to create any chosen superposition. (**Bottom right**) Potential-energy wells with barrier height lowered by applying a fast flux pulse for qubit state measurement.

to the image "slices" that are captured and combined into a three-dimensional picture in tomographic medical imaging) to characterize the quantum state. In particular, such measurements are used to reconstruct the density matrix of the system, a mathematical tool that specifies the components of an arbitrary quantum state. On page 1423 of this issue, Steffen *et al*. (*8*) report an important advance in the first tomographic measurements of an entangled state produced by two coupled solid-state qubits.

Steffen *et al*. use two superconducting phase qubits, A and B, coupled by a capacitance $C_x$ (see the figure, top panel). Each qubit consists of a Josephson tunnel junction (indicated by the X)—shunted with a capacitance $C$—in a superconducting loop of inductance $L$. The dynamics of the system are described by the motion of a fictitious particle representing the quantum variable δ, the difference between the phases of the superconducting order parameters on each side of the junction. This particle is confined to an asymmetric double-well potential $U(\delta)$ formed by applying an external magnetic flux (see the figure, bottom left panel). The two lowest energy levels in the shallow potential well on the left are the quantum states $|0\rangle$ and $|1\rangle$, separated by energy $E_{01}$. Fast microwave pulses at frequency $\nu_{01} = E_{01}/h$ (where $h$ is the Planck constant) prepare any chosen superposition of $|0\rangle$ and $|1\rangle$ (transitions from $|1\rangle$ to $|2\rangle$ can be ignored because their energy difference is off-resonance). Once state preparation is complete, a fast flux pulse tilts the potential (see the figure, bottom right panel). If the qubit is in the state $|1\rangle$, the phase particle tunnels to the adjacent deep potential well, causing a sudden change in δ and inducing a magnetic flux that is stored in the loop. If the qubit is initially in the state $|0\rangle$, however, no tunneling occurs. The difference between these two flux states is readily detected with an on-chip SQUID (superconducting quantum interference device) inductively coupled to the qubit.

In the case of two coupled phase qubits, there are four basis states: $|00\rangle$, $|01\rangle$, $|10\rangle$, and $|11\rangle$ (where 0 and 1 indicate the state of each individual qubit). Steffen *et al*. prepare the entangled state $(|01\rangle - i|10|)/\sqrt{2}$ (where $i = \sqrt{-1}$), one of the states that is important in quantum logic. The density operator for this state is $\hat{\rho} = (|01\rangle - i|10\rangle)(\langle 01| + i\langle 10|)/2$. The corresponding density matrix is

$$\rho = \begin{array}{c} \\ \langle 00| \\ \langle 01| \\ \langle 10| \\ \langle 11| \end{array} \begin{array}{c} \begin{array}{cccc} |00\rangle & |01\rangle & |10\rangle & |11\rangle \end{array} \\ \begin{pmatrix} 0 & 0 & 0 & 0 \\ 0 & 1/2 & -i/2 & 0 \\ 0 & i/2 & 1/2 & 0 \\ 0 & 0 & 0 & 0 \end{pmatrix} \end{array} \quad (1)$$

Entanglement is indicated by the nonzero, off-diagonal elements of the density matrix, $i/2$ and $-i/2$; these particular off-diagonal matrix elements must be nonzero to represent an entangled state. If one instead had a product state of the form, $(|10\rangle + |00\rangle)/\sqrt{2} = (|1\rangle + |0\rangle)|0\rangle/\sqrt{2}$, there would be no quantum correlations between measurements of the states of the two qubits. Simply measuring qubit A, for example, cannot distinguish between the entangled and product states described above, and each measurement would yield a 50% probability of being in $|0\rangle$ or $|1\rangle$. A more sophisticated sequence, namely, state tomography, is needed to determine all the elements of the density matrix.

Arguably, George Stokes (*9*) introduced such a procedure in 1852 in the context of linear optics. Using a set of four measurements involving polarizers of various orientations, he reconstructed the polarization state of an unknown electromagnetic wave. In the case of coupled phase qubits, a tomographic measurement involves applying different microwave pulse sequences (similar to those in nuclear magnetic resonance) before readout to obtain different linear combinations of the elements of the density matrix (*10*). From this information, Steffen *et al*. reconstruct the density matrix. Their results convincingly show the signatures of their entangled state, namely, the diagonal and nonzero off-diagonal matrix elements shown in Eq. 1. After correction for known measurement errors, the observed magnitudes are 87% of the theoretical values. The remaining discrepancy is consistent with predictions based on the measured decoherence time.

These tomographic measurements are a positive step forward for solid-state quantum computing, representing a proof-of-principle demonstration of the basic functions needed for a quantum computer. At the same time, we are reminded of the complexities of the solid state, which has many possible channels of decoherence. Fidelity—control and measurement precision—may be lost to uncontrolled degrees of freedom that might be associated with the readout circuit, low-frequency noise in charge, flux, and junction critical current, and lossy circuit materials. Steffen *et al*. suggest that their observed loss of fidelity is a result of poor dielectric materials. Decoherence in other kinds of superconducting qubits is reduced by operating them at symmetry points at which they are insensitive to environmental noise (*3*), thereby implementing a level of hardware fault tolerance. Moreover, quantum error correction codes have been developed for software fault tolerance. Given the tremendous progress made with superconducting qubits in the past few years, we expect the demonstration of even more sophisticated quantum algorithms in the not-too-distant future.

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# Detecting Awareness in the Vegetative State

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The vegetative state is one of the least understood and most ethically troublesome conditions in modern medicine. The term describes a unique disorder in which patients who emerge from coma appear to be awake but show no signs of awareness. Although the diagnosis depends crucially on there being no reproducible evidence of purposeful behavior in response to external stimulation (*1*), recent functional neuroimaging studies have suggested that "islands" of preserved brain function may exist in a small percentage of patients who have been diagnosed as vegetative (*2*). On this basis, we hypothesized that this technique also may provide a means for detecting conscious awareness in patients who are assumed to be vegetative yet retain cognitive abilities that have evaded detection using standard clinical methods.

In July 2005, a 23-year-old woman sustained a severe traumatic brain injury as a result of a road traffic accident. Five months later, she remained unresponsive with preserved sleep-wake cycles. Clinical assessment by a multidisciplinary team concluded that she fulfilled all of the criteria for a diagnosis of vegetative state according to international guidelines (*1*) [Supporting Online Material (SOM) text].

We used functional magnetic resonance imaging (fMRI) to measure her neural responses during the presentation of spoken sentences (e.g., "There was milk and sugar in his coffee"), which were compared with responses to acoustically matched noise sequences (*3*). Speech-specific activity was observed bilaterally in the middle and superior temporal gyri, equivalent to that observed in healthy volunteers listening to the same stimuli (fig. S1). Furthermore, sentences that contained ambiguous words (italicized) (e.g., "The *creak* came from a *beam* in the *ceiling*") produced an additional significant response in a left inferior frontal region, similar to that observed for normal volunteers. This increased activity for ambiguous sentences reflects the operation of semantic processes that are critical for speech comprehension.

An appropriate neural response to the meaning of spoken sentences, although suggestive, is not unequivocal evidence that a person is consciously aware. For example, many studies of implicit learning and priming, as well as studies of learning during anesthesia and sleep, have demonstrated that aspects of human cognition, including speech perception and semantic processing, can go on in the absence of conscious awareness.

To address this question of conscious awareness, we conducted a second fMRI study during which the patient was given spoken instructions to perform two mental imagery tasks at specific points during the scan (*3*). One task involved imagining playing a game of tennis and the other involved imagining visiting all of the rooms of her house, starting from the front door. During the periods that she was asked to imagine playing tennis, significant activity was observed in the supplementary motor area (Fig. 1). In contrast, when she was asked to imagine walking through her home, significant activity was observed in the parahippocampal gyrus, the posterior parietal cortex, and the lateral premotor cortex (Fig. 1). Her neural responses were indistinguishable from those observed in healthy volunteers (fig. S2) performing the same imagery tasks in the scanner (SOM text).

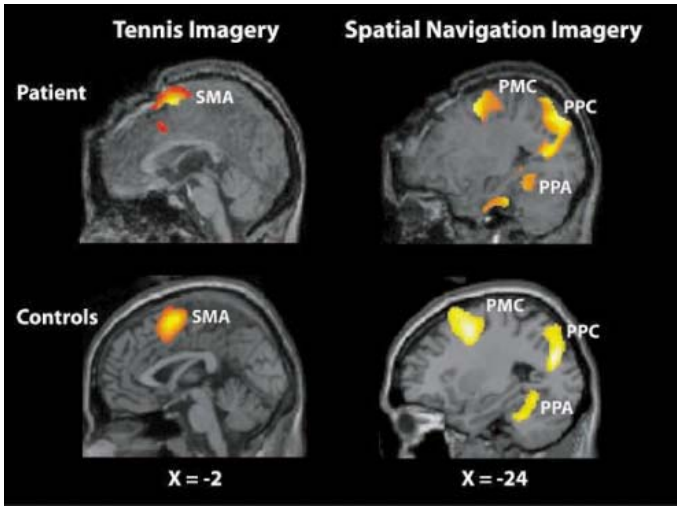


**Fig. 1.** We observed supplementary motor area (SMA) activity during tennis imagery in the patient and a group of 12 healthy volunteers (controls). We detected parahippocampal gyrus (PPA), posterior parietal-lobe (PPC), and lateral premotor cortex (PMC) activity while the patient and the same group of volunteers imagined moving around a house. All results are thesholded at $P < 0.05$ corrected for multiple comparisons. *X* values refer to distance in mm from the midline in stereotaxic space (SOM text).

These results confirm that, despite fulfilling the clinical criteria for a diagnosis of vegetative state, this patient retained the ability to understand spoken commands and to respond to them through her brain activity, rather than through speech or movement. Moreover, her decision to cooperate with the authors by imagining particular tasks when asked to do so represents a clear act of intention, which confirmed beyond any doubt that she was consciously aware of herself and her surroundings. Of course, negative findings in such patients cannot be used as evidence for lack of awareness, because false negative findings in functional neuroimaging studies are common, even in healthy volunteers. However, in the case described here, the presence of reproducible and robust task-dependent responses to command without the need for any practice or training suggests a method by which some noncommunicative patients, including those diagnosed as vegetative, minimally conscious, or locked in, may be able to use their residual cognitive capabilities to communicate their thoughts to those around them by modulating their own neural activity.

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3. Materials and methods are available as supporting material on *Science* Online.
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### Supporting Online Material

www.sciencemag.org/cgi/content/full/313/5792/1402/DC1
Materials and Methods
SOM Text
Figs. S1 and S2
References

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# Two Years at Meridiani Planum: Results from the Opportunity Rover

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The Mars Exploration Rover Opportunity has spent more than 2 years exploring Meridiani Planum, traveling ~8 kilometers and detecting features that reveal ancient environmental conditions. These include well-developed festoon (trough) cross-lamination formed in flowing liquid water, strata with smaller and more abundant hematite-rich concretions than those seen previously, possible relict "hopper crystals" that might reflect the formation of halite, thick weathering rinds on rock surfaces, resistant fracture fills, and networks of polygonal fractures likely caused by dehydration of sulfate salts. Chemical variations with depth show that the siliciclastic fraction of outcrop rock has undergone substantial chemical alteration from a precursor basaltic composition. Observations from microscopic to orbital scales indicate that ancient Meridiani once had abundant acidic groundwater, arid and oxidizing surface conditions, and occasional liquid flow on the surface.

The Mars Exploration Rover Opportunity landed at Meridiani Planum on 24 January 2004. During its 90-sol (martian solar day) nominal mission (*1*), Opportunity explored Eagle crater and the surrounding plains, studying laminated sulfate-rich sandstones that contain abundant hematite-rich spherules (*2*). Opportunity then spent nearly a year exploring Endurance crater, investigating ~7 m of exposed stratigraphic section in a unit dubbed the Burns formation (*3*). Observations there revealed chemical and textural changes with depth (*4*, *5*) and a stratigraphic section dominated by eolian dune and sand sheet facies but with evidence for subaqueous deposition in the uppermost half meter (*6*). Since leaving Endurance, Opportunity has traveled nearly 5 km to the south, assessing horizontal and vertical variations in geology over more regional scales. Major landmarks along the route (fig. S1) include Vostok crater, about 1.5 km south of Endurance, and Erebus crater, about 4 km south of Endurance. Pausing during the traverse, Opportunity also spent considerable time at the Olympia outcrop, just north of Erebus. Despite the generally homogeneous appearance of Meridiani Planum from orbit, a number of new features and phenomena have been observed at these locations at rover scale. Here, we present geologic results obtained along the traverse, interpretations of textural and geochemical observations at Endurance crater, and a refined model for the formation of the sedimentary rocks at Meridiani Planum.

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**Depositional processes.** Rocks of the Burns formation are finely stratified sandstones. By itself, this observation could be explained in terms of eolian or subaqueous sedimentation, accumulation by explosive volcanism, or emplacement as impact ejecta. On the basis of sedimentary structures and facies associations at Eagle (*2*) and Endurance (*6*) craters, we identified eolian and subaqueous sedimentation as the most probable deposition mechanisms.

It is well known that fine- to medium-grained sand, when subjected to shear stresses created by shallow, subaqueous flows with moderate current velocities, spontaneously forms highly sinuous-crested ripples with amplitudes of up to a few cm (*7*, *8*). In cross sections oriented transverse to flow, exposed laminae have a trough-shaped or "festoon" geometry (*9*). At the Olympia outcrop, Panoramic Camera (Pancam) (*10*) images of the rock Overgaard show a planar-laminated unit overlain by the best example of cm-scale trough cross-lamination found by Opportunity to date (Fig. 1A). These trough cross-laminae scour down into the underlying planar laminated unit from left to right. The trough cross-lamination forms a bed set with at least 3 to 4 cm of apparent thickness. Several superimposed sets show basal scouring and backfilling by concave- to occasionally convex-upward cross-laminae, which show a cross-cutting relation in which each trough-shaped subset truncates the subset to its left. In addition, the cross-strata exhibit a small angle of climb from left to right. Truncation surfaces and backfilling of subjacent sets are well expressed in the left-center part of the upper unit. Individual troughs are 3 to 4 cm wide. This particular type of cross-stratification can be further described as scalloped cross-lamination, which can form either by migration of a single bedform during fluctuating flow or by migration of superimposed bedforms in steady flow (*11*). We cannot distinguish between these two possibilities because of the lack of a complementary cut oriented parallel to the bedding plane.

A mosaic of Microscopic Imager (MI) (*12*) images was acquired to provide improved resolution of the Overgaard trough cross-lamination (Fig. 1B). The subgrain-scale resolution of this image provides additional supporting detail, including clear truncation of laminae that reinforces the Pancam observations. Stratal truncations of this sort are the result of primary discontinuities in sedimentation and cannot be artifacts produced by the intersection of bedding with topography. Moreover, fine-scale topography derived from stereoscopic MI imaging reveals a nearly flat surface for the portion of the outcrop that contains the trough cross-lamination. Because there is no systematic relationship of bedding to local slopes, this observation confirms that the cross-lamination is a primary attribute of the rock.

Cross-lamination of this type and at this scale is known to form subaqueously but is not known to develop in other types of flows, including eolian and volcanic base surges (*13*–*15*). We therefore discount suggestions that the Meridiani sandstones might have been formed as a part of volcanic (*16*) or impact (*17*) base surges. Furthermore, the overall stratigraphy and the succession of facies observed at Endurance crater are inconsistent with base surge deposits (*6*). The latter are characterized by flow deceleration sequences similar to subaqueous turbidites (*15*), and these have not been observed in the bedrock at Meridiani. Recent observations thus confirm the earlier interpretation of subaqueous transport and suggest that water flow was more extensive across the ancient Meridiani surface than was evident from observations at Eagle and Endurance craters alone.

Microscopic images provide additional information for testing depositional models. All outcrops imaged to date by Opportunity consist of medium-size sand grains that are rounded and well sorted. For example, Cobble Hill, a target within the interpreted eolian sand sheet facies in Endurance crater, has a mean grain size of 450 μm, with a standard deviation of 170 μm ($N$ = 200) (Fig. 2A). Walker (*18*) and Sparks (*19*) compiled grain-size distribution data for more than 300 pyroclastic flow deposits on Earth. The median grain size in Meridiani outcrops falls within the range of

medians recorded for terrestrial samples, but the sorting is distinctly different (*20*). In contrast, the size distribution of Meridiani sand grains falls well within the range of mean, median, and sorting values for terrestrial eolian sands (*21*).

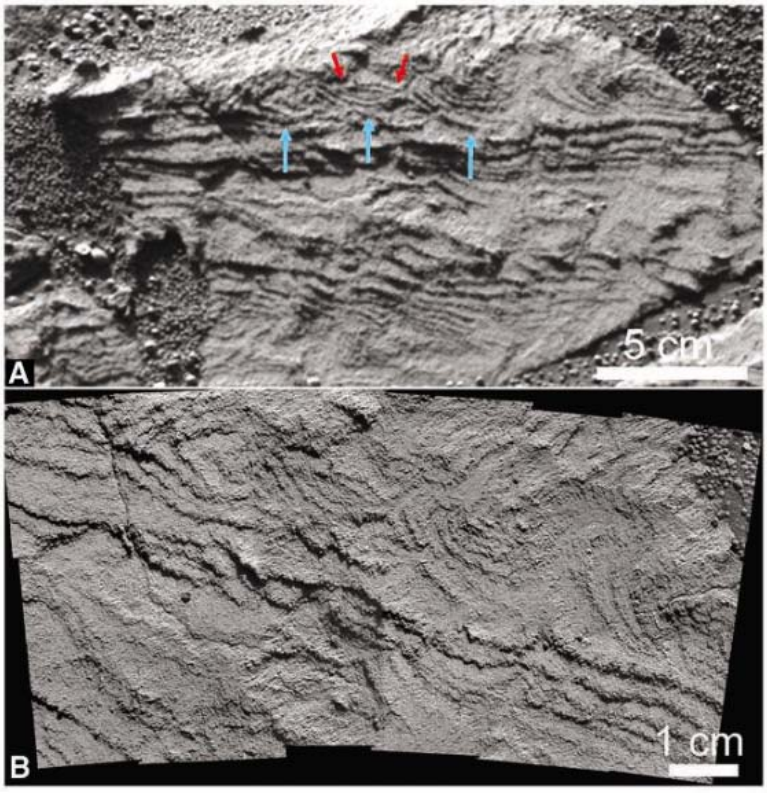


**Fig. 1.** Festoon (trough) cross-lamination in Overgaard. (**A**) Blue arrows point to three distinct festoons, indicated by basal truncation and concave-upward geometry. Red arrows point to downlapping, convex-up laminae that backfilled the festoon trough. (**B**) MI mosaic of part of Overgaard, showing generally granular texture and excellent sorting, right-to-left pinch out of planar-laminated unit, and stratal truncation and downlap of overlying festoon cross-laminae [compare with (A)]. Illumination is from the top in both images.

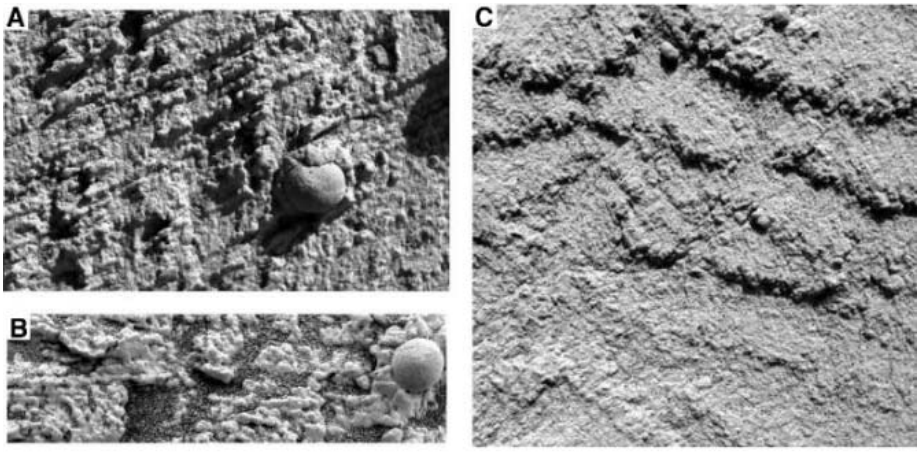


**Fig. 2.** Microscopic images of Meridiani outcrop rocks. (**A**) Rounded sand grains forming single-grain-thick laminae in target Cobble Hill, an interpreted eolian sand sheet within the measured stratigraphic section in Endurance crater. (**B**) Recrystallization textures in Eagle crater. At center left, a set of three or four laminae show fusing of grains. At right, the complete loss of primary fabric and development of blocky, interlocking crystals around a hematitic spherule implies a highly soluble precursor mineral assemblage. (**C**) Overgaard. Note the absence of visible spherules. Scale across each image is 3 cm.

This characteristic persists south of Endurance crater. Textures in Overgaard, and also in Strawberry, another outcrop near the rim of Erebus crater, again indicate only well-sorted, medium-grained sandstones. Grain-size distributions in sandstones near Erebus approximate those in Eagle and Endurance outcrops (Fig. 2C). Thus, outcrop-level sedimentary structures and fine-scale textures of Meridiani rocks both support the interpretation that they are eolian deposits, reworked locally by surface water.

**Geochemistry and mineralogy.** After using Opportunity's Rock Abrasion Tool (RAT) (*22*) to expose fresh rock surfaces, we collected Alpha Particle X-Ray Spectrometer (APXS) (*23*) data at Endurance crater that revealed systematic stratigraphic variations in chemical composition (*4*). From bottom to top of the measured stratigraphic section, Burns formation sandstones show decreasing amounts of Si, Al, Na, and K and increasing amounts of Mg and S. This trend is consistent with variations in the ratio of siliciclastic components to sulfates, with the fraction of siliciclastics decreasing upsection. The variations also correlate with diagenetic textural changes, including increased cementation and recrystallization and enhanced secondary porosity deeper in the section (*5*), and accordingly are inferred to result from postdepositional dissolution and mobilization of soluble salts (*4*).

Analysis of these chemical variations sheds light on the nature of the siliciclastic component (Fig. 3). The data form a curvilinear trend with samples from higher in the stratigraphic section plotting at lower $Al_2O_3/(FeO_T + MgO + CaO)$. The two mixing lines in the figure each have one endmember defined by the average composition of "chemical constituents," including sulfate salts, jarosite, and hematite, as inferred from Mössbauer spectroscopy (*24*) and chemical mass balance (*4*, *5*). The deviation of the data from any mixing line, like the lower one, that has unaltered martian basalt as the other endmember shows that the siliciclastic component in the current outcrop cannot be unaltered basalt.

The chemically altered basalt endmember composition for the upper mixing line was derived by assuming that roughly half of the divalent cations were removed from the precursor basalt during chemical weathering, enriching Al in the siliciclastic residue relative to Mg, Fe, and Ca. Deriving it in this way simulates the most important geochemical processes causing Al enrichment relative to divalent cations during chemical weathering of basalt: mineral dissolution and the precipitation of aluminosilicate phases. This mixing line coincides very closely to the Burns formation data. The best fit is found when ~55% of the total Mg, Fe, and Ca is removed (mostly Mg) and is consistent with the siliciclastic component being derived from chemical alteration of a precursor basalt. Further support for the idea

that the siliciclastic component is highly altered comes from Mini–Thermal Emission Spectra (TES) (*25*) infrared data, which show no pyroxene or olivine in the outcrop. Instead, a major component (~25%) of Al-rich amorphous silica is the dominant outcrop spectral signature along with sulfates.

It has been suggested on the basis of limited data from Eagle crater that simple sulfur addition (as $H_2SO_4$) to basalt with subsequent isochemical alteration might explain the chemistry and mineralogy of the Burns formation (*16*). This explanation would be consistent with geochemical data if all the points were clustered at a single location in Fig. 3, but the trend at Endurance crater rules it out. A mixing line involving addition of sulfur to basalt is a horizontal line on Fig. 3, clearly inconsistent with the trend. Addition of S to basalt could account for the trend only if the primary basaltic sediment composition varied considerably and systematically through the 7 m of stratigraphy exposed in Endurance crater and if the amount of S added varied directly with the primary basaltic composition, an implausible combination of coincidences.

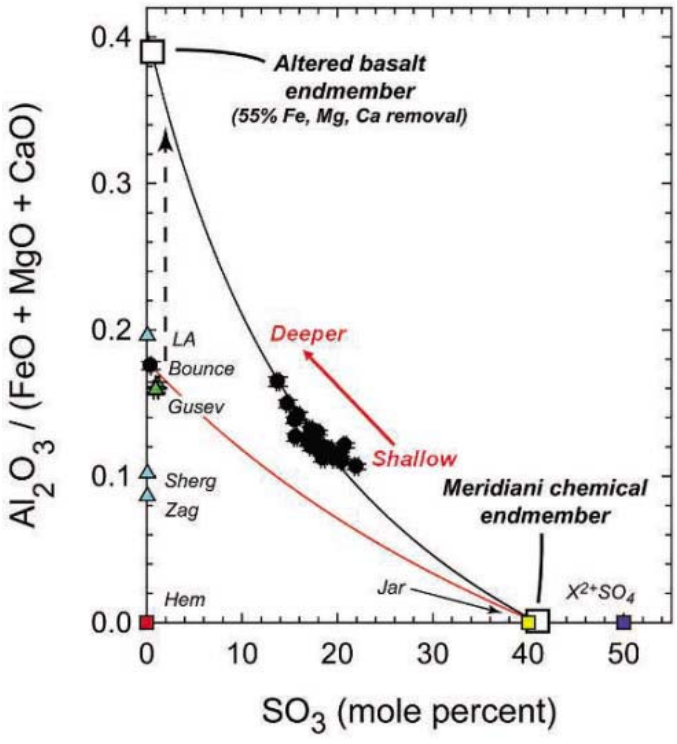


**Fig. 3.** Plot of molar $Al_2O_3/(FeO_T + MgO + CaO)$ versus $SO_3$ for the Burns formation (circles). [We sum the three major divalent cations to capture as much of the relevant geochemistry as possible in a single diagram, but we note that most of the observed variability in $Al_2O_3/(FeO_T + MgO + CaO)$ is due to variations in $Al_2O_3$ and MgO (*4*).] Also plotted are the compositions of martian basalts ("Zag," "Sherg," and "LA" denote the basaltic shergottite meteorites Zagami, Shergotty, and Los Angeles, respectively; "Bounce" denotes Bounce Rock, a piece of impact ejecta at Meridiani Planum (*1*); and "Gusev" denotes plains basalts at Gusev crater), the compositions of minerals observed within the Burns formation ("Hem" denotes hematite and "Jar" denotes jarosite), and a composition derived by removing 50% of the divalent cations (Ca, Mg, and Fe) from typical martian basalt to illustrate the effects of chemical weathering. $SO_3$ decreases and $Al_2O_3/(FeO_T + MgO + CaO)$ increases systematically with depth (red arrow), indicating an increasing proportion of the siliciclastic component with depth. Also shown are two mixing lines, one between average chemical constituents and unaltered basalt and a second between average constituents and altered basalt.

**Diagenesis.** Textural evidence for a fluctuating groundwater table in the Burns formation includes (*2*, *5*, *6*) (i) syndepositional sediment deformation, (ii) intergranular cement, (iii) mm-scale spherules interpreted to be concretions, (iv) mm-scale tabular pores interpreted to be crystal molds, (v) late generations of cement and recrystallization surrounding concretions (Fig. 2B), and (vi) decimeter-scale stratigraphically controlled zones of enhanced secondary porosity and recrystallization. As Opportunity traversed south, some features have changed and new textures have been observed.

At Eagle and Endurance craters, crystal molds represent the former presence of mineral grains with solubility comparable to magnesium sulfates, because they dissolved to form secondary porosity without seriously disrupting primary fabrics. Monoclinic habits suggest minerals such as melanterite ($Fe^{2+}SO_4 \cdot 7H_2O$) or calcium, iron, or magnesium chlorides.

North of Erebus crater, the rock Lemon Rind contains pseudomorphs after a mineral with an apparent cubic crystal habit (Fig. 4A). Millimeter-scale square, rectangular, and triangular shapes may suggest the former presence of "hopper crystals" commonly produced by halite in terrestrial evaporites (*26*, *27*). Hopper textures develop in cubic crystals because of preferred growth at edges and corners. They form in many evaporative settings (for example, brine-atmosphere and sediment-brine interfaces and capillary zones), but isolated hoppers within sediment suggest displacive growth above a groundwater capillary fringe (*26*, *28*), similar to the setting inferred for monoclinic-shaped crystal mold minerals (*5*).

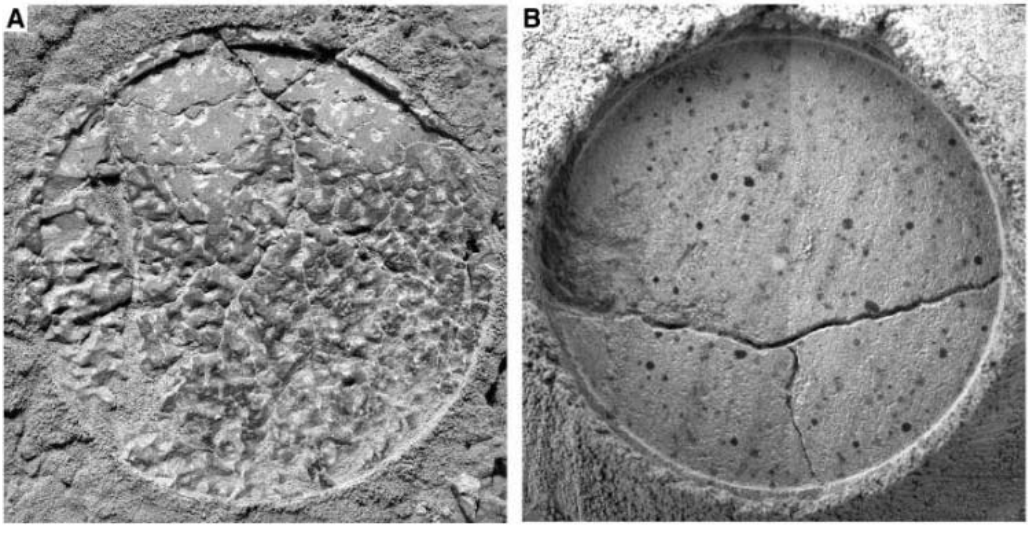


**Fig. 4.** MI mosaics of RAT holes that reveal diagenetic features observed only south of Endurance crater. (**A**) Apparent pseudomorphs after a mineral with a cubic crystal structure, perhaps halite, in the target Lemon Rind (see Fig. 5A for context). (**B**) Small, irregularly shaped hematite spherules in the target Kalavrita, at the Olympia outcrop. This RAT hole has also exposed a fracture intersection that is part of a polygonal fracture network. Scale across each image is 5 cm.

Within Eagle and Endurance craters, hematitic spherules are of uniform size, shape, and abundance and are interpreted to be sedimentary concretions formed in a near-isotropic groundwater flow regime (*2*, *5*). Spherules are characterized by a volume distribution that is more uniform than random, absence at bedding or other erosional surfaces, multiply fused spherules, and a distinctive composition (>50% $Fe_2O_3$ as hematite, with Ni/Fe far too low for a meteoritic origin). Neither accretionary lapilli nor oxidized metallic iron impact spherules, which might be other ways of creating small spherules (*17*), would plausibly have these characteristics.

As Opportunity traversed southward from Vostok crater to Olympia, the concretions became notably smaller (≤2 mm), more numerous, and more irregular in shape (Fig. 4B). The increase in number and decrease in size of concretions could reflect an increase in the nucleation rate of hematite or its precursor. Nucleation rates of iron oxides are strongly controlled by the degree of supersaturation with respect to the precipitating mineral phase (*29*, *30*), so an increase in the number of concretions could result from a sudden generation of supersaturated conditions caused by changes in the chemistry of the diagenetic fluid. South of Olympia, adjacent to Erebus crater, concretions are absent at the resolution of the MI (Fig. 2C), but this appears to be a relatively local, possibly facies-controlled, phenomenon because small concretions reappear in outcrops south of Erebus.

**Later modification.** Three types of later modification are particularly prominent in the Burns formation (Fig. 5): surface rinds up to several millimeters thick that record surface alteration, erosionally resistant fracture fills that include a cement component, and networks of polygonal fractures.

Rinds differ chemically from subjacent outcrop, notably showing enrichment of Na and Cl and depletion of S. They are particularly well developed where they have formed at the interface between outcrop surfaces and thin coverings of soils. This observation suggests that the rind formation process may be ubiquitous and ongoing on exposed or thinly covered outcrop surfaces, but that its rate is substantially slower than the eolian sandblasting of soft outcrop rock by saltating grains that is pervasive across the Meridiani plains. In this model, only where rock surfaces have been protected from sandblasting, for example, when buried by a thin veneer of soil and only recently uncovered, is thick rind formation observed.

Fracture fills are erosionally resistant, often vertically oriented features associated with linear fractures of possible impact origin. These features are spectrally distinct from adjacent outcrop but differ chemically only in detail. APXS data indicate that fracture fills contain siliciclastic materials in amounts similar to or slightly greater than nearby outcrop lithologies; the fill is typically slightly enriched in Al and Si and depleted in Mg and S. Unlike rinds, fracture fills show no substantial Na or Cl enrichment. The high abundance of silicates means that the fills are not primarily precipitated, but the absence of basaltic minerals indicates that fractures are not filled by present-day soils. Instead, the close similarity of fracture fill and country rock lithologies suggests that fractures were filled primarily by intraclastic material derived from adjacent outcrops. The limited total volume of alteration rinds and fracture fill indicates very low aggregate rates of fluid flow and chemical weathering during the time since the Meridiani outcrop rocks were deposited.

Polygonal crack systems commonly mark Meridiani outcrop rocks (Figs. 4B and 5, A and B). These three-dimensional crack systems have typical polygon dimensions of ~10 cm and have been observed both on loose boulders within Endurance crater and on exposed bedrock surfaces south of Endurance. Fracture networks on rock surfaces commonly cut across bedding, indicating that the fracturing significantly postdates the deposition process. Because sulfate minerals can undergo substantial volume reduction upon dehydration, the fractures likely resulted from volume reduction during water loss from hydrated sulfate minerals as environmental conditions became more desiccating.

**Temporal relations.** Key events in the history of Meridiani Planum include the formation of sulfate-rich sand grains by acid sulfate alteration of basalt, deposition of sand grains to form laminated rock, emplacement of the hematite-rich spherules, and other diagenetic events.

Several lines of evidence demonstrate that sulfate-rich sand grains formed before deposition of the outcrop rock seen today. Perhaps the most compelling of these is the compositional gradient at Endurance crater. The observed vertical gradient in the ratio of sulfates to siliciclastics is readily explained by interaction of a sulfate-rich sandstone with groundwater, but it cannot plausibly be explained by any process in which unaltered basaltic sand grains first form laminated outcrop rock and then are chemically altered in place with no groundwater interaction. Also, acid sulfate alteration of the grains that form the outcrop rocks is pervasive along the rover's full traverse, with no less-altered or unaltered zones anywhere, consistent with substantial reworking and mixing after the alteration but unlikely for alteration in place by vapor or small amounts of water.

Additional evidence comes from textural observations. Many hematitic concretions have been sectioned by the RAT and then imaged by the MI, and none preserves relict sand textures in its interior at 30 μm/pixel resolution. This observation requires that sandstone textures within the concretions were obliterated during concretion growth, consistent with sand grains made of soluble sulfates and siliciclastic materials much finer-grained than the sands themselves, but not with sand grains made of basalt. Also, the grains in the Meridiani outcrop are larger and better rounded than windblown basaltic sands on the current surface of Meridiani Planum. The simplest explanation is that the outcrop grains were made of lower-density, less-resistant materials at the time of their transport and emplacement. Geological and geochemical observations thus agree that the acid sulfate alteration of parent basalts invoked to explain Meridiani chemistry took place in an as-yet undiscovered setting before the accumulation of Meridiani sands now observed in outcrop.

It is also clear that the hematitic spherules formed after the acid sulfate alteration of the

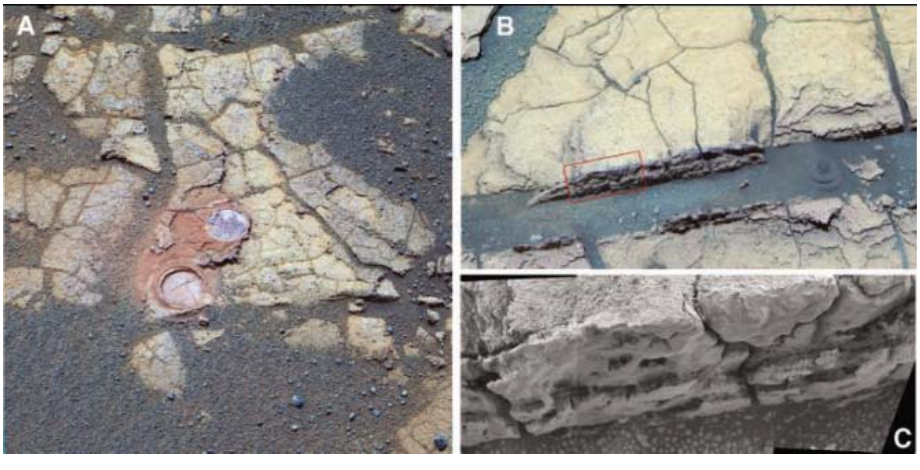


**Fig. 5.** Evidence of late modification of Meridiani outcrop rocks. (**A**) Pancam false color image of dark rinds atop outcrop rock at Fruit Basket, just north of Erebus crater; round features are RAT holes 4.5 cm in diameter in the rind (upper, Lemon Rind) and subjacent lithology (lower, Strawberry). (**B**) Pancam false-color image of Roosevelt fracture fill along the margin of a linear fracture at Erebus crater. (**C**) Partial MI mosaic ~4 cm wide of the Roosevelt fracture fill, showing mm-scale lamination within the fill; approximate location is shown in red box in (B). Note also the polygonal fracturing of rocks in (A) and (B).

sand grains took place. Sand grains in Meridiani deposits readily recrystallized around spherules (Fig. 2B), which is likely if the grains were sulfate-rich at the time of spherule formation but inconsistent with the sand grains having had a basaltic lithology at that time.

By comparison to eolian deposits on Earth, facies development of the measured section in Endurance crater is interpreted to reflect the interaction between eolian processes and a migrating groundwater table (*6*), with water-deposited beds near the top of the section reflecting aqueous reworking of sands in low-lying areas where ground waters reached the surface. Thus, groundwater infiltration must have begun as Meridiani sands accumulated, not later. The systematic increase in sulfate content observed from the bottom to the top of the measured section may reflect this penecontemporaneous infiltration.

As documented by petrographic textures in MI images, diagenesis ensued after formation of the sand grains and their deposition to form the outcrop: Precipitation of lamina-cutting monoclinic crystals preceded the growth of hematite-rich concretions, which in turn came before the recrystallization of grains, the precipitation of cements around concretions, and the formation of secondary porosity, including the dissolution of earlier formed diagenetic crystals. Diagenesis was largely completed by the time that Meridiani's current geomorphic surface formed; the limited amount of later modification (e.g., rinds, fracture fills, and polygonal fractures) indicates that water has been scarce in the Meridiani region for the past several billion years.

**Regional relations.** The sulfate-rich deposits examined by Opportunity are located at the top of a several-hundred-meter-thick section of rock that disconformably covers the underlying Noachian cratered terrain (*31*). In fact, the hematite-bearing plains surface materials encountered by Opportunity are dominated by an eolian cover of basaltic sands and lag deposits of hematitic concretions left behind as wind deflated the relatively soft sulfate-rich bedrock. Further, rock exposures to the east, north, and west of the hematite-bearing plains form the etched terrains that extend over several hundred thousand square kilometers (*31*, *32*). These etched-terrain bedrock materials are layered and highly eroded by wind. Analyses of Mars Express OMEGA (Observatoire pour la Mineralogie, l'Eau, les Glaces et l'Activité) data show that the etched terrains have an enhanced 1.92-μm absorption band relative to the Noachian cratered terrain, indicating enhanced abundances of water-bearing mineral phases (*33*). In some areas, kieserite and perhaps polyhydrated sulfate minerals have been identified in etched terrain exposures (*33*, *34*). Thus, Opportunity has examined the top of a widely exposed section of layered deposits that are hydrated and demonstrably sulfate-bearing in multiple locations. This information allows us to consider models for formation of the layered deposits that place rover-based observations into a regional context.

**Summary.** The first stage in the development of the rocks observed by Opportunity was acid sulfate alteration of basaltic source material to produce sand grains composed of Mg, Ca, and Fe sulfates mixed with a very fine-grained siliciclastic residue. These grains were then reworked by wind in the presence of fluctuating groundwater that occasionally came to the surface and flowed across it. Interaction with substantial amounts of groundwater produced hematite-bearing concretions and a variety of other diagenetic textures, as well as a subsurface gradient in composition at Endurance crater.

None of Opportunity's observations to date reveal the environment in which the sulfate-rich sand grains originally formed. Given the compelling evidence for emergence of groundwaters at Meridiani under generally arid climate conditions, we suggest that the most likely mechanism is that grains originated by erosion from a dirty playa, a pan of sulfate precipitates and fine-grained siliciclastic particles formed by interaction of precursor basalts with acidic groundwaters, followed by evaporation (*2*, *5*, *6*).

In the absence of deposits that have not been reworked by wind and water, other mechanisms for formation of the sand grains, including acid sulfate weathering in a volcanic environment [a component of the scenario suggested by McCollom and Hynek (*16*)], cannot be ruled out. Whatever the formation mechanism, however, it is clear that the grains now observed in outcrop were emplaced by eolian and aqueous processes, and that after their emplacement they interacted with substantial quantities of groundwater.

Although we cannot pinpoint the location where the sand grains formed, we note that there is no need to invoke transport to Meridiani from a distant source region. The eolian and aqueous processes that produced the observed sedimentary facies could have operated exclusively on local scales, so it is plausible that the sulfate-rich sand grains formed at Meridiani, rather than having been transported from elsewhere.

Whatever process produced the sulfate-rich sands at Meridiani, it created enough material to cover several hundred thousand square kilometers. Layered deposits of sulfate minerals have also been found in a number of other areas across the martian surface (*33*). In contrast, hematite has been detected only in a few places on Mars, of which Meridiani is the largest. Because it is the formation of an erosional veneer that renders Merdiani hematite visible from orbit, it may be that other hematite accumulations have thus far escaped detection. But it is also possible that other regions of Mars lacked the specific conditions for groundwater diagenesis responsible for hematite precipitation at Meridiani (*35*).

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## Supporting Online Material

www.sciencemag.org/cgi/content/full/313/5792/1403/DC1
Fig. S1

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# *Hoxa2*- and Rhombomere-Dependent Development of the Mouse Facial Somatosensory Map

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In the mouse trigeminal pathway, sensory inputs from distinct facial structures, such as whiskers or lower jaw and lip, are topographically mapped onto the somatosensory cortex through relay stations in the thalamus and hindbrain. In the developing hindbrain, the mechanisms generating such maps remain elusive. We found that in the principal sensory nucleus, the whisker-related map is contributed by rhombomere 3–derived neurons, whereas the rhombomere 2–derived progeny supply the lower jaw and lip representation. Moreover, early *Hoxa2* expression in neuroepithelium prevents the trigeminal nerve from ectopically projecting to the cerebellum, whereas late expression in the principal sensory nucleus promotes selective arborization of whisker-related afferents and topographic connectivity to the thalamus. *Hoxa2* inactivation further results in the absence of whisker-related maps in the postnatal brain. Thus, *Hoxa2*- and rhombomere 3–dependent cues determine the whisker area map and are required for the assembly of the whisker-to-barrel somatosensory circuit.

The rodent trigeminal pathway represents a suitable system to study neuronal connectivity and pattern formation (*1*–*5*). Somatosensory inputs from distinct facial regions are collected through the mandibular (supplying the lower jaw and lip), the maxillary (supplying the whiskers and the upper jaw and lip), and the ophthalmic branches of the trigeminal nerve by first-order neurons whose cell bodies reside in the trigeminal ganglion (Fig. 1). The central processes of trigeminal ganglion neurons enter the hindbrain and give ascending and descending branches wiring into the rostral principal (PrV) and the caudal spinal (SpV) nuclei. Sensory inputs are in turn transmitted to the somatosensory cortex through a relay station in the ventral posterior medial (VPM) nucleus of the thalamus. At all levels of the pathway, the spatial arrangements of neurons and their afferent fibers faithfully reproduce the physical distribution of peripheral sensory receptors generating somatotopic facial representations. However, little is known about the mechanisms underlying the development of somatotopy.

At early developmental stages, the vertebrate hindbrain is transiently segmented into rhombomeres (r) along the anteroposterior axis (*6*). Fate-mapping studies revealed that the progeny of individual segments form compact transverse stripes of cells running throughout the ventriculopial axis of the postnatal hindbrain (*7*). However, the relationship between the early segmental plan and the establishment of topographical circuitry in the developing hindbrain is still poorly understood. At the molecular level, *Hox* genes are crucial determinants of rhombomere identity and neuronal patterning (*8*–*11*). One possibility is that, at later stages of hindbrain maturation, *Hox* expression may set up a molecular program for somatotopic map formation. Here, we focused on *Hoxa2*, the most rostrally expressed *Hox* gene in the hindbrain, and on the development of somatotopy in the PrV nucleus, which has a fundamental role in thalamic and cortical pattern formation (*12*, *13*). We show that cellular segregation of rhombomere progenies and maintenance of *Hoxa2* expression in PrV through later stages of hindbrain development provide a set of positional labels that instructs both the wiring of peripheral trigeminal afferents and the topographic axonal mapping of PrV target neurons to VPM thalamus (summary in fig. S1).

**Rhombomere-dependent somatotopy of PrV.** The somatotopic organization of the rodent trigeminal system can be conveniently visualized by cytochrome oxidase (CO) histochemistry (Fig. 1, A, D, G, and J) (*5*, *14*–*16*). At postnatal day 4 (P4), the dorsal component of the PrV nucleus appears as a lightly CO-stained area that includes the lower jaw and lip representation (Fig. 1, A and G) and receives inputs from the mandibular branch of the trigeminal nerve. The ventral PrV component is instead organized in neuronal modules, or barrelettes, replicating the array of whiskers and sinus hairs on the snout (Fig. 1, A and G) that are innervated by the maxillary branch. Similar neuronal arrangements also exist at thalamic and cortical levels both for the lower jaw and lip and for whisker-related representations (known as barreloids and barrels, respectively) (Fig. 1, D and J). However, little is known about the cellular mechanisms underlying the establishment of interareal somatotopy during prenatal development, resulting in segregation of the lower jaw and lip and the whisker-related maps.

Here, we asked whether PrV somatotopy relates to its rhombomeric origin. To permanently label rhombomere progenies, we mated mouse lines expressing the Cre recombinase selectively in r2 (*R2::Cre*) (*17*) or in r3 and r5 (*Krox20::Cre*) (*18*) with the Z/AP reporter line (*19*). In this mating scheme, Cre-mediated recombination results in permanent activation of alkaline phosphatase (AP) in rhombomere progenies. AP and CO stainings on adjacent cross-sections at P4 revealed that, in the PrV, the territory including the barrelettes and sinus hair–related neuronal clusters is contributed by the r3 progeny (r3p) (Fig. 1, G to I). In contrast, the unpatterned CO-stained portion of PrV, containing the map of the lower jaw and lip, is composed entirely by the r2 progeny (r2p) and anteriorly delimited by the r1 progeny (r1p) (Fig. 1, A to C and fig. S2, A to D). Posteriorly, the barrelette field is bordered by the r4 progeny (r4p) (Fig. 1, G to I and fig. S2, E to H). Notably, the progeny of r5 (r5p) did not contribute to the PrV nucleus, nor to the more posteriorly located interpolaris (SpVi) or caudalis (SpVc) subdivisions of the SpV column (Fig. 1, G to I and fig. S2), the only other neuronal formations generating whisker-related representations (*16*).

**Rhombomere-related topography of PrV axonal projections.** Next, we mapped the AP-stained axonal projections from the brainstem onto the CO-stained representations of distinct face areas in the VPM on adjacent sections. In *Krox20::Cre;Z/AP* animals at P4, AP-stained axon terminals precisely matched the CO-stained barreloids and sinus hair–related neuronal modules in the dorsolateral VPM (Fig. 1, J to L). Most of these projections to VPM originated from r3-derived PrV neurons, given that the r5 progeny did not contribute to any of the nuclei generating the main stream of ascending whisker-related afferents to VPM—i.e., PrV, SpVi, or SpVc (*20*) (Fig. 1 and fig. S2). Notably, in *R2::Cre;Z/AP* animals the AP-stained projections adopted instead a complementary pattern. Axon terminals from r2-derived PrV neurons mainly segregated into the ventromedial VPM, containing the representation of the lower jaw and lip, and were excluded from the barreloid area (Fig. 1, D to F).

**Rhombomere-specific patterns of trigeminal primary afferent arborization.** The mapping data suggested that r3-derived PrV neurons predominantly receive inputs from the whiskers, supplied by the maxillary division of the trigeminal nerve, whereas inputs from the mandibular division are relayed through the r2-derived portion of

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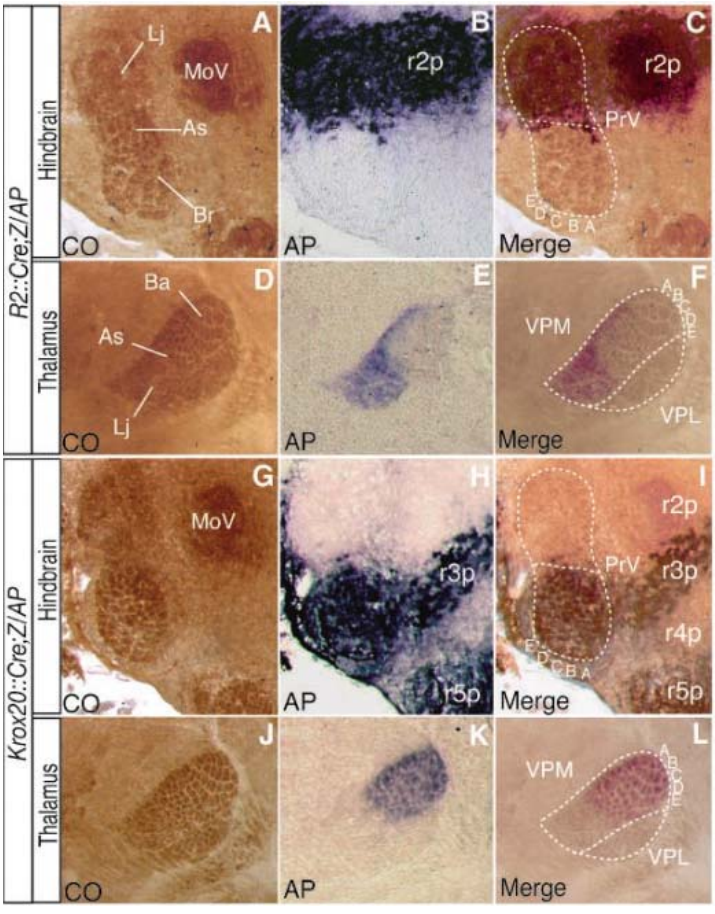
PrV. To test this possibility, we assessed the central projection patterns of trigeminal primary afferents (Figs. 2 and 3).

By embryonic day 10.5 (E10.5), the trigeminal nerve has already entered the hindbrain through an entry point in r2 (*10*, *21*), and ascending axons are arrested shortly thereafter at their final position. By fluorescent dextran tracing of trigeminal afferents and enhanced green fluorescent protein (EGFP) detection in the hindbrain of $Hoxa2^{EGFP}$ embryos (*22*), we mapped the point of arrest next to the r1/r2 border (Fig. 3D). By E14.5, somatotopy of the trigeminal tract was evident, as determined by labeling individual branches with distinct fluorescent dextrans (Fig. 2, A to C). Specifically, mandibular axons were located dorsally, ophthalmic axons ventrally, and maxillary afferents run in-between the two other divisions. Notably, the relative width of each division within the nerve varied along the rostrocaudal axis (Fig. 2, A to C), corresponding to differential patterns of arborization into the PrV target nucleus (Fig. 2, D to F). Caudally to the nerve entry point, the maxillary axons and their radially oriented collaterals contributed to most of the dorsoventral extent of the tract (in red, Fig. 2, A to C and E), with very little input from mandibular (in green, Fig. 2, A, C, E, and F) or ophthalmic (in violet, Fig. 2, B and C) divisions. In contrast, mandibular collaterals rostral to the nerve entry point were conspicuous and spatially segregated from maxillary collaterals (Fig. 2, A, D, and F); moreover, no collaterals could be identified from the ophthalmic branch (Fig. 2, B and C).

Such rostrocaudal variations suggested that the arborization patterns from mandibular or maxillary afferents may be related to the rhombomeric origin of PrV target neurons. To correlate arborization patterns with rhombomeric domains, we used the $Hoxa2^{EGFP(lox–neo–lox)}$ allele (*22*) that expresses EGFP only upon Cre-mediated deletion of the selection marker cassette, thus allowing tracing of rhombomere-specific domains of $Hoxa2^{EGFP}$-expressing cells by mating with suitable Cre-expressing mouse lines. Therefore, we simultaneously visualized collaterals from individually labeled trigeminal branches and r2- or r3-derived *Hoxa2*-expressing territories by EGFP detection in E14.5 $R2::Cre;Hoxa2^{EGFP(lox–neo–lox)}$ (Fig. 2, G and K) or $Krox20::Cre;Hoxa2^{EGFP(lox–neo–lox)}$ (Fig. 2, L and N) fetuses, respectively. Notably, the entire dorsoventral extent of the r3-derived portion of PrV was selectively targeted by collaterals from maxillary axons (arrows, Fig. 2, J and M; summaries in Fig. 2, K and N), with very little, if any, mandibular input (Fig. 2, I, K, L, and N). In contrast, mandibular axons selectively sent collaterals dorsally within the r2-derived portion of PrV (arrows, Fig. 2G; summary in Fig. 2H). Thus, individual trigeminal divisions differentially contribute to the innervation of PrV in relation to the rhombomeric origin of target neurons, with the r3-derived PrV neurons receiving input almost uniquely from the maxillary division.

***Hoxa2* differential expression in PrV.** At early stages, *Hoxa2* is expressed throughout r2 and r3, although at different levels (*10*). At E14.5, in situ hybridization in comparison with the PrV-specific marker *Drg11* (*23*) revealed restricted expression of *Hoxa2* in developing PrV neurons (Fig. 2, P and Q) but not in trigeminal ganglion cells. Comparison of transcript distribution and AP staining on adjacent sections from *Krox20::Cre;Z/AP* fetuses re-



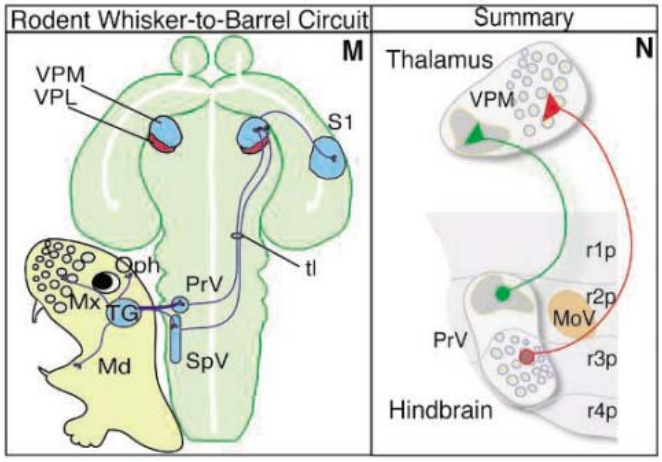




**Fig. 1.** Rhombomere-related somatotopic organization and connectivity of PrV nucleus. (**A** to **L**) Cross-sections through hindbrain PrV [(A) to (C) and (G) to (I)] and thalamic VPM [(D) to (F) and (J) to (L)] nuclei in P4 *R2::Cre;Z/AP* [(A) to (F)] or *Krox20::Cre;Z/AP* [(G) to (L)] animals, respectively. Face maps are revealed by cytochrome oxidase (CO) histochemistry at PrV [(A) and (G)] and VPM [(D) and (J)] levels. Lower jaw– (Lj), anterior snout (As) sensory hair–, and whisker-related neuronal patterns are shown. Whisker-related neuronal modules (Br, barrelettes; Ba, barreloids) are organized into five rows, labeled A through E. The progenies of r2 (r2p) (B) or r3 and r5 (r3p and r5p) (H) are traced by alkaline phosphatase (AP) staining, and superimposed (Merge) to CO-stained adjacent sections in (A) or (G), respectively. At PrV level, r2p (C) or r3p (I) specifically contribute to the Lj or the As and Br maps, respectively. At VPM level, the Lj map is entirely targeted by AP-stained axon terminals from r2-derived neurons [(E) and (F)], whereas axon terminals from *Krox20::Cre ;Z/AP* animals selectively target As and Ba neuronal modules, precisely matching CO-stained neuronal modules [(K) and (L)]. (**M**) Diagram of trigeminal circuit in mouse. (**N**) Summary showing the relationship between rhombomere progenies and somatotopy of PrV nucleus and its axonal connections to VPM. Md, mandibular branch of trigeminal nerve; MoV, trigeminal motor nucleus; Mx, maxillary branch; Oph, ophthalmic branch; PrV, principal sensory trigeminal; S1, somatosensory cortex; SpV, spinal sensory trigeminal column; TG, trigeminal ganglion; tl, trigeminal lemniscus; VPL, ventroposterior lateral nucleus; VPM, ventral posterior medial nucleus.

vealed that the r3-derived portion of PrV expressed high levels of *Hoxa2* (compare Fig. 2, O and P). In contrast, *Hoxa2* expression was much lower in r2p, specifically in the dorsal part of PrV (Fig. 2P). Such transcript distributions correlated with the r2- or r3-specific arborization patterns of mandibular or maxillary afferents, respectively, suggesting that high levels of *Hoxa2* expression may be involved in selective wiring of the maxillary division.

**Early *Hoxa2* requirement in r2 for trigeminal nerve pathfinding.** To address *Hoxa2* function, we analyzed the patterns of trigeminal afferents in $Hoxa2^{EGFP-/-}$ homozygous mutants. A notable pathfinding defect was observed in E16.5 (Fig. 3, A and B) and E14.5 (Fig. 3, E, F, H, and I) homozygous mutants. The ascending branch of the trigeminal nerve did not stop to encapsulate the PrV nucleus, but ectopically projected to the cerebellum. Labeling of homozygous mutants at E10.5, before PrV nucleus formation (*24*), revealed that the pathfinding defect was already present at this early stage (arrows, Fig. 3, D and G).

To address the *Hoxa2* spatial requirement, we next deleted *Hoxa2* selectively in r2 by mating *R2::Cre* transgenic mice with our $Hoxa2^{flox}$ allele (*17*). Notably, in *R2::Cre;*$Hoxa2^{flox/flox}$ fetuses the trigeminal tract also ectopically projected to the cerebellum (Fig. 3C), similarly to the full knockout phenotype. Thus, the normal arrest of trigeminal afferents in the rostral hindbrain does not require information from PrV target neurons, but it is dependent on early expression of *Hoxa2* in the r2 neuroepithelium.

**Late *Hoxa2* requirement for maxillary axon arborization.** To investigate *Hoxa2* involvement during arborization of trigeminal afferents, we used an approach based on tamoxifen (TM) inducible *CMV–*β*actin–Cre-ERT2* (*CMV::Cre–ERT2*)/*loxP* to achieve time-dependent inactivation of *Hoxa2* (*25*). Given that arborization of trigeminal afferents into PrV neurons started at about E13 (*26*), *CMV::Cre-ERT2;*$Hoxa2^{flox/flox}$ fetuses and $Hoxa2^{flox/flox}$ controls were chronically administered with TM for three times at E12.5, E13.0, and E13.5. Fetuses were collected at E14.5 and in situ hybridization confirmed no or little residual *Hoxa2* expression in the hindbrain of TM-treated *CMV::Cre-ERT2;*$Hoxa2^{flox/flox}$ mutants (*25*). Retrograde labeling of individual trigeminal branches revealed a selective inhibition of collateral formation from the maxillary branch (Fig. 3, J, L, M, N, P, and Q). Moreover, some collaterals from the mandibular division appeared to be abnormally oriented toward the territory normally targeted by maxillary collaterals (arrow, Fig. 3O; summary in Fig. 3Q), suggesting a degree of competitive interactions between trigeminal branches (*27*). In contrast, we did not observe in these animals ectopic trigeminal projections to the cerebellum, confirming that such a phenotype depended on *Hoxa2* function earlier than E12.5.

To further support a *Hoxa2* role for maxillary axon arborization, we generated *Krox20::Cre;*$Hoxa2^{flox/flox}$ fetuses. At E14.5, we observed a severe reduction of maxillary collaterals into the r3-derived PrV (Fig. 3, R, T, and U), with occasional ectopic reorientation of mandibular collaterals, similar to the TM-induced knockout animals (Fig. 3, O, Q, S, and U). Thus, *Hoxa2* is required in PrV target neurons to induce selective arborization of maxillary axons.

**Loss of *EphA4* and *EphA7* expression in PrV of *Hoxa2* mutants.** In E15.5 *Krox20::Cre;*$Hoxa2^{flox/flox}$ homozygous mutant fetuses, normal specification and organization of PrV neurons

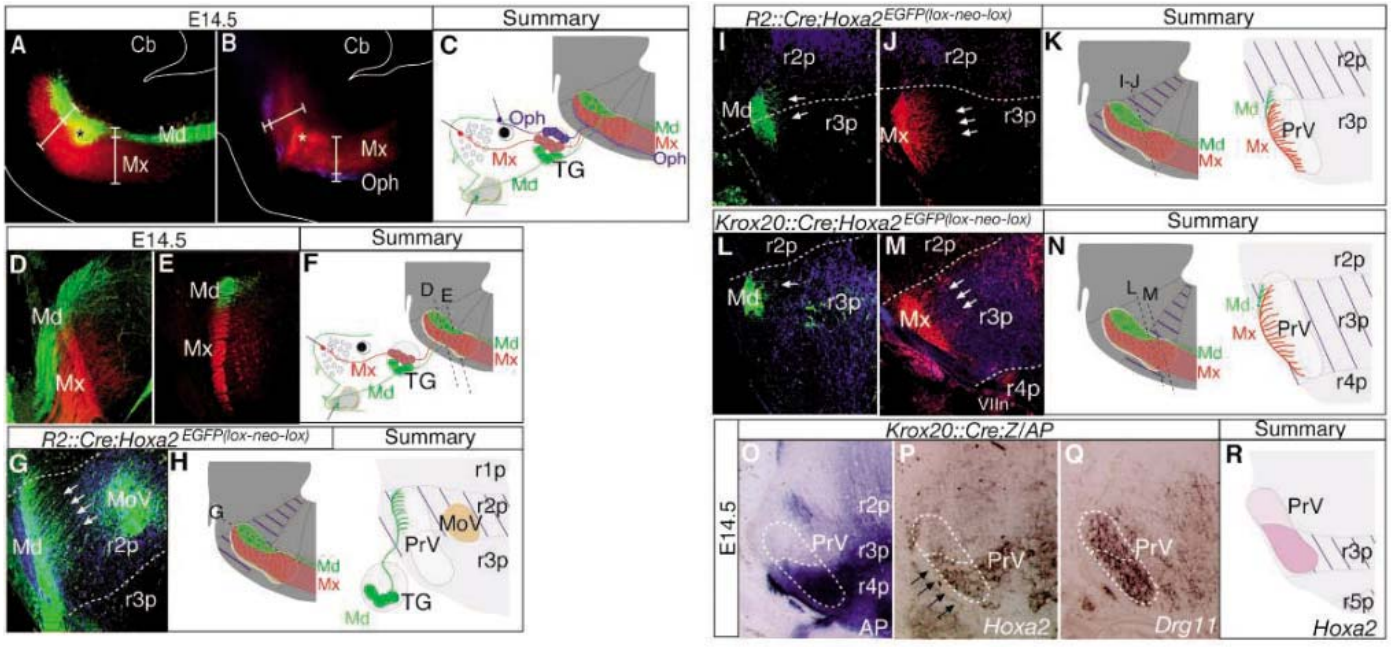



**Fig. 2.** Rhombomere-specific arborization patterns of trigeminal afferents and *Hoxa2* expression. (**A** to **C**) Trigeminal tract somatotopy in whole-mount E14.5 wild-type brain. Maxillary (Mx) and mandibular (Md) (A), or Mx and ophthalmic (Oph) (B) branches were simultaneously retrogradely labeled by rhodamin-dextran [red in (A) and (B)] or Alexa 488-dextran [green in (A) and violet in (B)], respectively. Crosses in summary diagram (C) indicate local patterns of arborization of axons. The Md branch runs dorsally, Oph ventrally, and Mx in between. The relative width of each branch varies along the rostrocaudal axis [bars in (A) and (B)]. The star (★) indicates the trigeminal nerve entry point. Cb, cerebellum. (**D** to **F**) Arborization patterns and spatial segregation of radially oriented collaterals from retrogradely labeled Md (green) and Mx (red) branches [(D) and (E)] in coronal sections through PrV nucleus at (D) and posterior (E) to the nerve entry point [as in summary (F)]. Posteriorly, the PrV is predominantly targeted by collaterals from Mx. (**G** to **N**) Rhombomere-specific patterns of arborization of Md and Mx branches. Md (green) or Mx (red) branch dextran labeling and simultaneous detection of EGFP (blue) from r2-derived (r2p) or r3-derived (r3p) progenies in *R2::Cre;*$Hoxa2^{EGFP(lox-neo-lox)}$ [(G), (I), and (J)] or *Krox20::Cre ;*$Hoxa2^{EGFP(lox-neo-lox)}$ [(L) and (M)] E14.5 fetuses, respectively. Selective arborization of Md branch in r2p [arrows, (G) and (I)], whereas no or very few collaterals are formed in r3p (L). [(J) and (M)] R3p is specifically targeted by Mx collaterals (arrows) [summary in (H), (K), and (N)]. (**O** to **Q**) [and summary in (**R**)] Coronal adjacent sections through PrV (outlined by dashed line) from *Krox20::Cre;Z/AP* E14.5 fetuses. AP staining in (O) visualizes r3p, whereas (P) and (Q) show in situ hybridization with *Hoxa2* and PrV-specific *Drg11* probes, respectively. High-level *Hoxa2* expression in r3p, but not in r2p, of PrV target neurons correlates with maxillary branch arborization.

was observed, as assessed by *Drg11* expression (fig. S3, A and D). However, *EphA4* and *EphA7* transcripts were lacking or severely reduced in the r3-derived portion of PrV, whereas *ephrinA5* expression in the target thalamus was unaffected (fig. S3, B, C, E, F, J, and K). Importantly, we also observed similar spatially restricted impairments of *EphA4* and *EphA7* expressions in the PrV of TM-treated *CMV::Cre-ERT2;Hoxa2*$^{flox/flox}$ mutant fetuses (fig. S3, H and I). These results confirmed a late requirement of *Hoxa2* and indicated that topographic wiring defects may be present in the mutants as a result of impaired *EphA* receptor expression.

**Absence of whisker-related maps and altered topography of PrV axonal connectivity in *Hoxa2* mutants.** At E16.5, the analysis of AP-stained axons in *Krox20::Cre;Hoxa2*$^{flox/flox}$*;Z/AP* mutants demonstrated normal pathfinding from PrV to VPM (fig. S4, A to F). However, CO-stainings at P4 revealed the absence of whisker-related neuronal patterns at both the PrV and VPM levels in these mutants (Fig. 4). Notably, analysis of AP-stained axon terminals revealed that, within the VPM, axons from PrV did not establish a normal topographic branching pattern and were mistargeted. Specifically, the majority of fibers did not project to the barreloid field, but ectopically targeted the VPM ventromedial region (Fig. 4, H, K, M, and N). Moreover, apoptotic loss of r3-derived PrV neurons was observed between P0 and P4 (Fig. 4, B and E, and fig. S5), which was not observed at prenatal stages (Fig. 4, A and D); this difference was likely the consequence of the partial deafferentation and miswiring of the circuit during the postnatal critical period of sensory experience (*28*). Thus, the inactivation of *Hoxa2* during prenatal development altered the wiring properties of PrV neurons with regard to both peripheral afferents and thalamic VPM target neurons, resulting in topographic changes of the trigeminal circuit in mutant postnatal brain (summarized in fig. S1).

**Conclusions.** We found that rhombomere-specific cellular cues are important for the spatial segregation of PrV neurons into a somatotopic pattern. Specifically, the r3 progeny contribute to the portion of the PrV that segregates into whisker-related neuronal patterns at postnatal stages. The persistence of rhombomere-specific cohesion properties of postmitotic progeny into late stages of hindbrain development may provide a cellular framework upon which to build precise neuronal connectivity. In addition, we demonstrated that *Hoxa2* has multiple spatiotemporal roles in the assembly of the trigeminal circuit. First, we demonstrated an early requirement for *Hoxa2* expression in the r2 neuroepithelium to prevent entering of peripheral afferents in r1. However, inner ear vestibular afferents, running just lateral to trigeminal axons, are not arrested at the r1/r2 border and normally project to the cerebellum (*29*). Thus, a *Hoxa2*-dependent molecular barrier may exist throughout r2 and/or at the r1/r2 border that is specifically involved in arresting the pathfinding of trigeminal but not vestibular axons.

Second, the onset of arborization of trigeminal axons into the PrV follows a rhombomere-specific pattern. Specifically, r3-derived neurons selectively receive collaterals from maxillary axons, whereas mandibular axons arborize predominantly into the r2-derived portion of PrV. Such a pattern correlates with differential levels and distribution of *Hoxa2* transcripts in the PrV nucleus: High or low expression domains corresponded to wiring by maxillary or mandibular axons, respectively. Moreover, spatiotemporally induced inactivations estab-

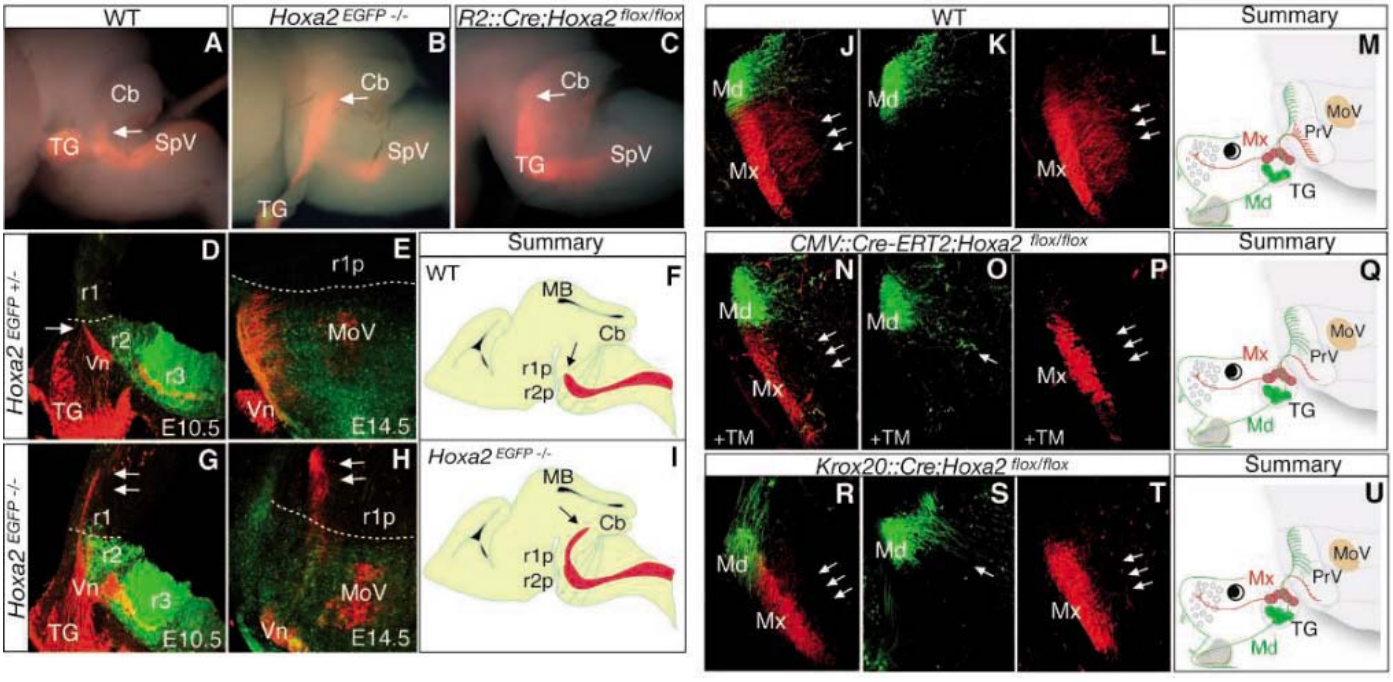



**Fig. 3.** Spatiotemporal requirement of *Hoxa2* for trigeminal afferent pathfinding and arborization. (**A** to **C**) Retrograde labeling of trigeminal nerve by whole trigeminal ganglion (TG) injection of rhodamine-conjugated dextran in E16.5 wild-type (WT) (A), *Hoxa2*$^{EGFP-/-}$ (B), and r2-specific knockout (*R2::Cre;Hoxa2*$^{flox/flox}$) (C) fetuses. In (B) and (C), the nerve ascending branch does not stop at its normal position after entering the hindbrain, but ectopically projects to the cerebellum. (**D** to **F**) Cross-sections through E10.5 (D) and E14.5 (E) *Hoxa2*$^{EGFP+/-}$ specimen doubly labeled for rhodamine and EGFP showing the entrance of the trigeminal nerve (Vn) tract at r2 level and its point of arrest at the r1/r2 border [summary drawing in (F)]. (**G** to **I**) Cross-sections through E10.5 (G) and E14.5 (H) *Hoxa2*$^{EGFP-/-}$ specimen. In homozygous mutants, Vn ectopically projects toward the cerebellar territory [summary in (I)]. These data demonstrate an early requirement for *Hoxa2* in r2 to arrest incoming ascending afferents. (**J** to **U**) Coronal sections through the hindbrain of E14.5 WT [(J) to (M)], tamoxifen (TM)–induced *CMV::Cre-ERT2;Hoxa2*$^{flox/flox}$ [(N) to (Q)], and r3/r5-specific *Krox20::Cre;Hoxa2*$^{flox/flox}$ [(R) to (U)] conditional homozygous mutant fetuses. Mandibular (Md) and maxillary (Mx) branches and their collaterals were simultaneously retrogradely labeled by Alexa 488-dextran (green) and rhodamin-dextran (red), respectively. Maxillary branch arborization is selectively inhibited both after *Hoxa2* temporal inactivation during collateral formation (TM administration at E12.5 through E13.5) [arrows, (P)] and spatial inactivation in r3 [arrows, (T)] [summaries in (Q) and (U)]. Reorientation of mandibular axon collaterals is also observed [arrows in (O) and (S)]. Specimens in (N) to (P) and (R) to (T) are distinct. Cb, cerebellum; MB, midbrain.

lished a specific requirement for *Hoxa2* during collateral formation from the maxillary division (Fig. 3). Notably, trigeminal ganglion neurons, which do not express *Hoxa2*, displayed a normal somatotopic organization in the mutants (fig. S6), supporting the idea that the topography of trigeminal peripheral processes is independent of central influences but that target cell maturation in the brainstem is an important regulator of arborization of central afferents (*26*). Thus, *Hoxa2* function in PrV neurons may be important to regulate the expression of molecules involved in trigeminal afferent arborization, such as neurotrophins and their receptors (*30*, *31*), Slit proteins and Robo receptors (*32*), and/or semaphorins and neuropilin receptors (*33*).

Third, our data indicate the involvement of *Hoxa2* in the topographic wiring of axonal connections to the VPM thalamus. While guidance of PrV axons to the thalamus was not affected in *Hoxa2* mutants, selective changes occurred in the topographic specificity of axonal mapping within the VPM nucleus. Graded expressions of ephrins and Eph receptors have been involved in sensory mapping (*34*–*37*). The finding that Hoxa2 positively regulates *EphA4* and *EphA7* expressions in the PrV indicates that Hoxa2-mediated control of connectivity could partly occur through Eph receptor function. By governing the distribution of molecules providing positional mapping labels, Hoxa2 could simultaneously regulate topographic wiring between brainstem (PrV) and thalamus (VPM), as well as between the periphery (incoming trigeminal afferents) and PrV. Moreover, as soluble homeobox proteins can function as guidance factors for axons (*38*), additional mechanisms might be at work to establish *Hoxa2*-mediated topographic wiring. Together with recent evidence indicating a role in motoneuron connectivity (*39*), these data begin to point to *Hox* genes as fundamental players in the building of sensorimotor circuitry in vertebrates.

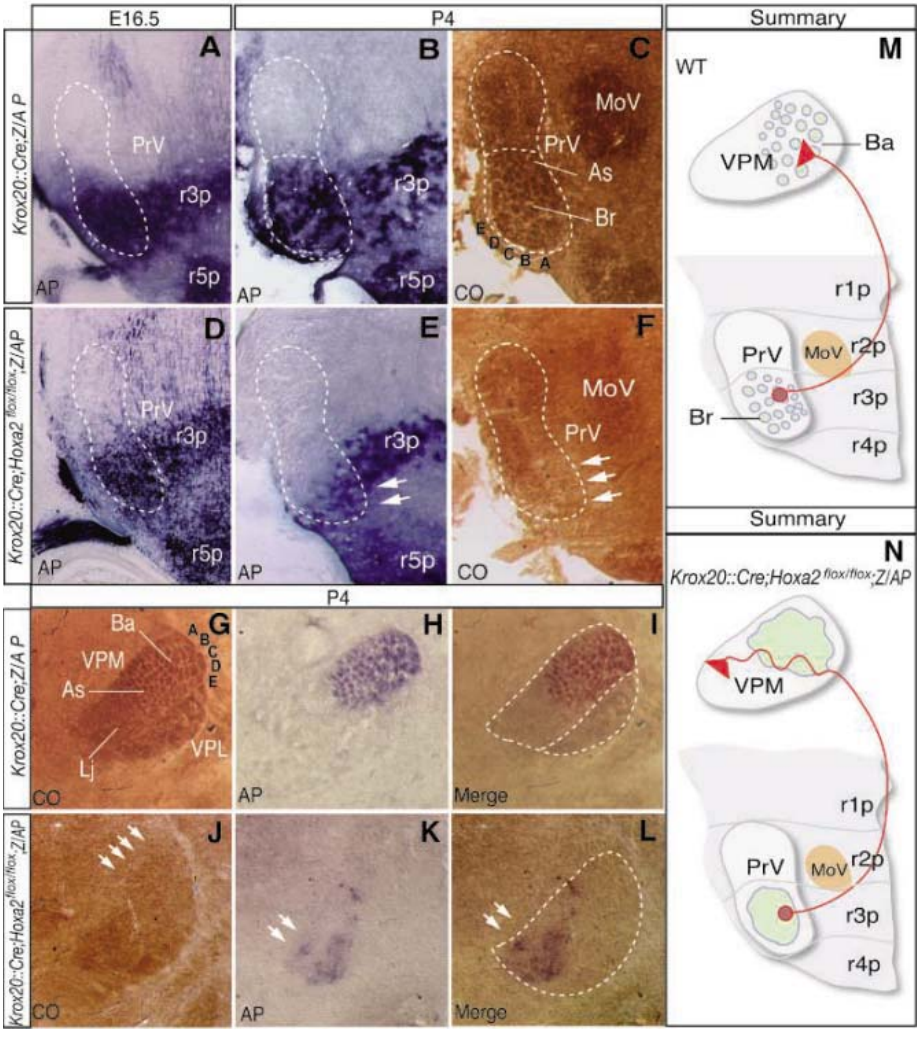



**Fig. 4.** Absence of whisker-related neuronal patterns and altered topography of PrV connectivity in *Hoxa2* mutants. (**A** to **F**) Cross-sections through PrV nucleus (outlined) of *Krox20::Cre;Z/AP* [(A) to (C)] and *Krox20::Cre;Hoxa2*$^{flox/flox}$*;Z/AP* homozygous mutants [(D) to (F)] at E16.5 [(A) and (D)] and P4 [(B), (C), (E), and (F)]. Cytochrome oxidase (CO) histochemistry reveals absence of barrelettes (Br) in mutant PrV at P4 (F), and progressive loss of alkaline phosphatase (AP)-stained r3-derived (r3p) PrV neurons between E16.5 (D) and P4 (E). (**G** to **L**) Cross-sections through the thalamus of *Krox20::Cre;Z/AP* [(G) to (I)] and *Krox20::Cre;Hoxa2* $^{flox/flox}$*;Z/AP* [(J) to (L)] P4 animals. [(G) and (J)] Ventroposterior medial (VPM) nuclei are visualized by CO, whereas AP stainings on adjacent sections [(H) and (K)] map axon terminals from the PrV neurons in panels [(B) and (E)]. Lack of barreloids (Ba) is observed in *Krox20::Cre;Hoxa2*$^{flox/flox}$*;Z/AP* mutant VPM [arrows, (J)], whereas AP staining demonstrates mistargeting of PrV axon terminals to ventromedial VPM [(K), and merge in (L)]. (**M** and **N**) Summary drawings.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**Supporting Online Material**
www.sciencemag.org/cgi/content/full/1130042/DC1
Materials and Methods
Figs. S1 to S6
References

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# Exotic Earths: Forming Habitable Worlds with Giant Planet Migration

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**Close-in giant planets (e.g., "hot Jupiters") are thought to form far from their host stars and migrate inward, through the terrestrial planet zone, via torques with a massive gaseous disk. Here we simulate terrestrial planet growth during and after giant planet migration. Several-Earth-mass planets also form interior to the migrating jovian planet, analogous to recently discovered "hot Earths." Very-water-rich, Earth-mass planets form from surviving material outside the giant planet's orbit, often in the habitable zone and with low orbital eccentricities. More than a third of the known systems of giant planets may harbor Earth-like planets.**

To date, giant planets have been detected around almost 200 main-sequence stars (*1*, *2*). An unexpected result is the abundance of planets very close to their host stars—about 40% of the known extrasolar planets are interior to Mercury's orbital distance of 0.4 astronomical units (AU; 1 AU is the Sun-Earth distance), although observational biases favor the detection of hot Jupiters (*3*). The occurrence of close-in giant planets is surprising because models predict that giant planets form much more easily in the cold, outer regions of protoplanetary disks (*4*, *5*). These planetary systems have been attributed to inward migration of a giant planet on $10^5$-year time scales caused by an imbalance of torques generated by the gaseous protoplanetary disk (*6*–*9*). In the process, the giant planet moves through the terrestrial planet zone (located from a few tenths of an AU to about 2 to 3 AU). Radioactive dating of solar system material (*10*) and observations of dust dispersal in disks around young stars (*11*) indicate that rapid precipitation and coagulation of solid material in the inner regions of circumstellar disks are likely, leading to the question of the fate of these protoplanets during and after giant planet migration. Previous studies on the possibility of Earth-like planets coexisting with close-in giant planets are divided (*12*–*16*).

Here, we simulate the growth and dynamical evolution of protoplanetary material from small bodies to terrestrial planets during and after the migration of a giant planet through the terrestrial zone (see supporting online material for details). Simulations start from a circumstellar disk in the middle stages of planet formation, extending from 0.25 to 10 AU. The disk contains 17 Earth masses ($M_\oplus$) of rocky/icy material, evenly divided between 80 Moon- to Mars-sized "planetary embryos" (*17*) and 1200 "planetesimals" with properties modified so that each body behaves as a collection of less massive objects (*18*). The disk has a compositional gradient: The inner disk is iron-rich and water-poor whereas the outer disk is water-rich and iron-poor [as in (*19*) but with 50% water by mass beyond 5 AU]. A Jupiter-mass giant planet starts at 5 AU and is migrated in to 0.25 AU in $10^5$ years (*8*). The orbits of all bodies in each simulation are integrated for 200 million years with the hybrid symplectic integrator Mercury (*20*), modified to include two additional effects: (i) "type 2" giant planet migration (*6*, *13*) and (ii) aerodynamic gas drag (*18*) from a gaseous disk that dissipates on a $10^7$-year time scale (*21*).

At early times (Fig. 1) the giant planet migrates inward through the disk, causing nearby material to either be scattered outward onto high-eccentricity orbits (*13*) or shepherded inward by the giant planet's moving mean-motion resonances (*22*). The buildup of inner material induces rapid growth of a 4 $M_\oplus$ planet just inside the 2:1 mean motion resonance in $10^5$ years [also shown by (*16*)]. Smaller bodies (planetesimals) feel a stronger drag force and are shepherded by higher-order resonances (in this case, the 8:1 resonance) and form a pileup of 0.2 $M_\oplus$ at 0.06 AU. At the end of the migration period, the remaining disk material is divided between bodies captured in low-eccentricity orbits in interior resonances with the Jupiter-mass planet and higher-eccentricity orbits beyond 0.5 AU. The protoplanetary disk is now dynamically hot (i.e., orbital eccentricities and inclinations are high), and accretion proceeds at a slower rate than would occur in a nonstirred, dynamically cold disk. However, the gas continues to damp eccentricities and inclinations, also causing the orbits of icy planetesimals from the outer disk to decay inward on million-year time scales, delivering a large amount of water to the growing terrestrial planets. After the gas dissipates (at $10^7$ years), the disk is

**Table 1.** Properties of simulated planets. Results are from four simulations (see supporting online material for details).

| | Hot Earths | Normal terrestrials | Hab. zone planets | Outer terrestrials | Solar system* |
|---|---|---|---|---|---|
| Mean number of planets | 0.25† | 2 | 0.5 | 11 | 4 |
| Mean planet mass ($M_\oplus$) | 4.2 | 1.1 | 2.0 | 0.6 | 0.49 |
| Mean water mass fraction | $2 \times 10^{-2}$ | $8 \times 10^{-2}$ | $8 \times 10^{-2}$ | $3.5 \times 10^{-1}$ | $4 \times 10^{-4}$ |
| Mean iron mass fraction | 0.25 | 0.28 | 0.27 | 0.14 | 0.32‡ |
| Mean orbital eccentricity | 0.01 | 0.23 | 0.10 | 0.23 | 0.08 |
| Mean orbital inclination (°) | 0.7 | 11 | 7 | 13 | 3.0 |

*Solar system physical properties are from (*37*) and orbital properties are from (*35*). †Every simulation with gas drag formed one to three hot Earths during giant planet migration. However, in many cases an artificial drag force caused continued inward migration of the giant planet. In these cases, the integrator usually introduced an error causing the eventual ejection of hot Earths when they entered ~0.05 AU. We therefore consider 0.5 a lower bound on the frequency of hot Earths in these systems. See also (*16*). ‡Solar system iron mass fractions are calculated without Mercury, because of its anomalously high iron content.

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stirred by interactions between bodies, and clearing continues through scattering. After 200 million years the inner disk is composed of the collection of planetesimals at 0.06 AU, a 4 $M_\oplus$ planet at 0.12 AU, the hot Jupiter at 0.21 AU, and a 3 $M_\oplus$ planet at 0.91 AU. Previous results have shown that these planets are likely to be stable for billion-year time scales (*15*). Many bodies remain in the outer disk, and accretion and ejection are ongoing due to long orbital time scales and high inclinations.

Two of the four simulations from Fig. 2 contain a >0.3 $M_\oplus$ planet on a low-eccentricity orbit in the habitable zone, where the temperature is adequate for water to exist as liquid on a planet's surface (*23*). We adopt 0.3 $M_\oplus$ as a lower limit for habitability, including long-term climate stabilization via plate tectonics (*24*). The surviving planets can be broken down into three categories: (i) hot Earth analogs interior to the giant planet; (ii) "normal" terrestrial planets between the giant planet and 2.5 AU; and (iii) outer planets beyond 2.5 AU, whose accretion has not completed by the end of the simulation. Properties of simulated planets are segregated (Table 1): hot Earths have very low eccentricities and inclinations and high masses because

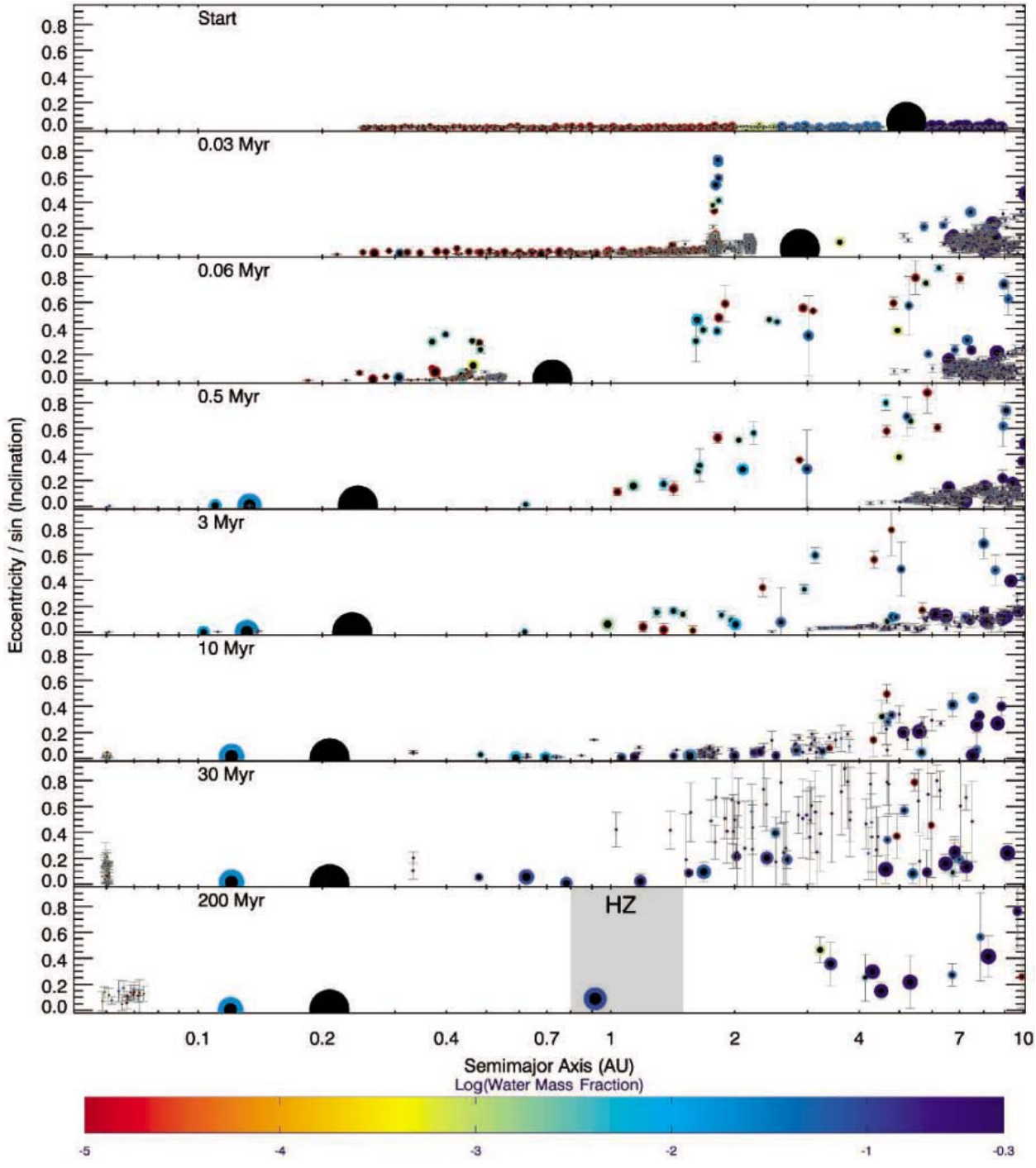


**Fig. 1.** Snapshots in time of the evolution of one simulation. Each panel plots the orbital eccentricity versus semimajor axis for each surviving body. The size of each body is proportional to its physical size (except for the giant planet, shown in black). The vertical "error bars" represent the sine of each body's inclination on the *y*-axis scale. The color of each dot corresponds to its water content (as per the color bar), and the dark inner dot represents the relative size of its iron core. For scale, the Earth's water content is roughly $10^{-3}$ (*28*).

they accrete on the migration time scale ($10^5$ years), so there is a large amount of damping during their formation. These planets are reminiscent of the recently discovered, close-in 7.5 $M_\oplus$ planet around GJ 876 (*25*), whose formation is also attributed to migrating resonances (*26*).

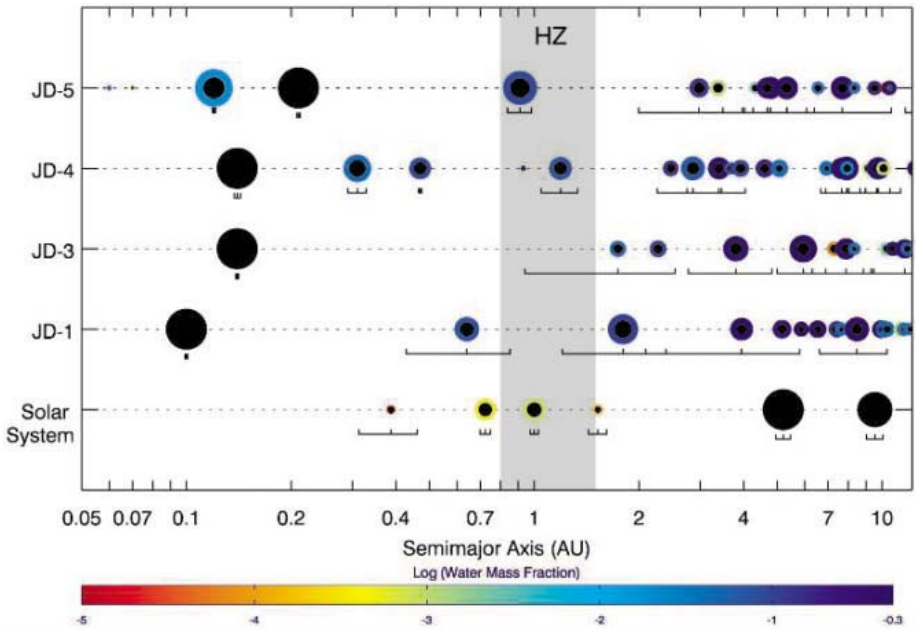


**Fig. 2.** Final configuration of our four simulations, with the solar system shown for scale. Each simulation is plotted on a horizontal line, and the size of each body represents its relative physical size (except for the giant planets, shown in black). The eccentricity of each body is shown beneath it, represented by its radial excursion over an orbit. As in Fig. 1, the color of each body corresponds to its water content, and the inner dark region to the relative size of its iron core. The simulation from Fig. 1 is JD-5. Orbital values are 1-million-year averages; solar system values are 3-million-year averages (*35*). See table S1 for details of simulation outcomes. Note that some giant planets underwent additional inward migration after the end of the forced migration, caused by an artificial drag force. This caused many hot Earths to be numerically ejected, but had little effect outside the inner giant planet. See supporting online material for details.

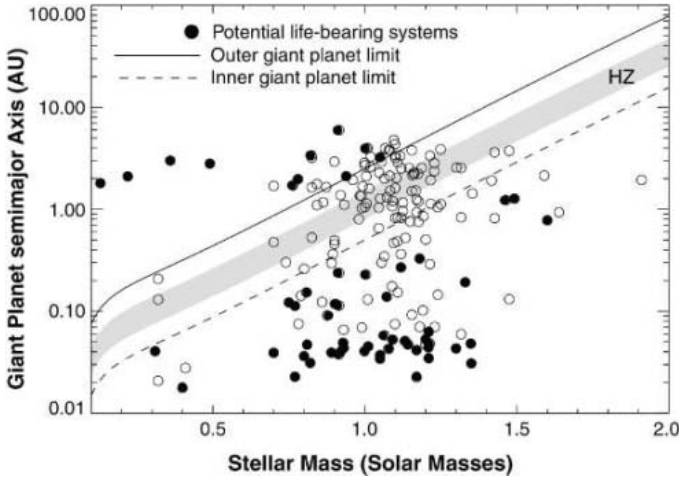


**Fig. 3.** Giant planet orbital parameter space that allows terrestrial planets to form in the habitable zone. The solid line indicates the limit for outer giant planets from (*30*). The dashed line is an approximate limit (0.5 AU with eccentricity less than 0.1—the maximum eccentricity achieved in most simulations—for a solar-mass star) inside which low-eccentricity giant planets allow for the formation of habitable planets, derived from our results and (*15*). We calculated the habitable zone (HZ, shaded area) by assuming the temperature to scale with the stellar flux (i.e., the square root of the stellar luminosity), using a stellar mass-luminosity relation fit to data of (*36*). Open circles represent known giant planets that are unlikely to allow habitable terrestrial planets in the habitable zone. Filled circles represent known planets with low enough orbital eccentricities to satisfy our criteria for habitable planet formation, deemed to be potentially life-bearing.

Farther from the star, accretion time scales are longer and the final phases take place after the dissipation of the gas disk (at $10^7$ years), causing the outer terrestrials to have large dynamical excitations and smaller masses, because accretion has not completed by 200 million years; collisions of outer bodies such as these may be responsible for dusty debris disks seen around intermediate-age stars (*27*). In the "normal" terrestrial zone, dynamical excitations and masses fall between the two extremes as planets form in a few times $10^7$ years, similar to the Earth's formation time scale (*10*). In addition, the average planet mass in the terrestrial zone is comparable to the Earth's mass, and orbital eccentricities are moderate (Table 1).

Both the hot Earths and outer Earth-like planets have very high water contents [up to >100 times that of Earth (*28*)] and low iron contents compared with our own terrestrial planets (Table 1). There are two sources for these trends in composition: (i) strong radial mixing induced by the migrating giant planet, and (ii) an influx of icy planetesimals from beyond 5 AU from gas drag-driven orbital decay that is unimpeded by the scattering that Jupiter performs in our own system. The outer terrestrial planets acquire water from both of these processes, but the close-in giant planet prevents in-spiraling icy planetesimals from reaching the hot Earths. The accretion of outer, water-rich material dilutes the high iron content of inner disk material, so water-rich bodies naturally tend to be iron-poor in terms of mass fraction. The high water contents of planets that formed in the habitable zone suggest that their surfaces would be most likely covered by global oceans several kilometers deep. Additionally, their low iron contents may have consequences for the evolution of atmospheric composition (*29*).

The spacing of planets (Fig. 2) is highly variable; in some cases planets form relatively close to the inner giant planet. The ratio of orbital periods of the innermost >0.3 $M_\oplus$ terrestrial planet to the close-in giant ranges from 3.3 to 43, with a mean (median) of 12 (9). We can therefore define a rough limit on the orbital distance of an inner giant planet that allows terrestrial planets to form in the habitable zone. For a terrestrial planet inside the outer edge of the habitable zone at 1.5 AU, the giant planet's orbit must be inside ~0.5 AU (the most optimistic case puts the giant planet at 0.68 AU). We apply this inner giant planet limit to the known sample of extrasolar giant planets [including planets discovered by the radial velocity, transit, and microlensing techniques (*1*, *2*)] in combination with a previous study of outer giant planets (*30*). We find that 54 out of 158 (34%) giant planetary systems in our sample permit an Earth-like planet of at least 0.3 $M_\oplus$ to form in the habitable zone (Fig. 3). The fraction of known systems that could be life-bearing may therefore be considerably higher than previous estimates (*30*).

The occurrence of hot Jupiters appears to be a strong function of stellar metallicity (*31*). In addition, the solid component of protoplanetary disks is assumed to be proportional to metallicity. Therefore, systems such as the ones studied here may have very massive solid disks and could have systematically larger planet masses. If, for example, such disks are more likely to form $\sim 10\ M_{\oplus}$ "hot Neptunes" [e.g., 55 Cnc e (*32*)] than $\sim 4\ M_{\oplus}$ hot Earths, then our disk is too small by a factor of a few. Assuming that planet mass scales with disk mass, the typical mass of a habitable planet in such systems may be several Earth masses. In addition, our calculations were for a realistic but fixed giant planet mass and migration rate. Less (more) massive giant planets or faster (slower) migration rates increase (decrease) the survival rate of terrestrial material exterior to the close-in giant planet (*13*).

Upcoming space missions such as the National Aeronautics and Space Administration's Kepler and Terrestrial Planet Finder and the European Space Agency's COROT and Darwin will discover and eventually characterize Earth-like planets around other stars. We predict that a large fraction of systems with a close-in giant planet will be found to have a hot Earth or potentially habitable, water-rich planets on stable orbits in the habitable zone. Suitable targets may be found in the known giant planet systems.

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# Observation of Electroluminescence and Photovoltaic Response in Ionic Junctions

**Daniel A. Bernards,[1] Samuel Flores-Torres,[2] Héctor D. Abruña,[2] George G. Malliaras[1]***

**Electronic devices primarily use electronic rather than ionic charge carriers. Using soft-contact lamination, we fabricated ionic junctions between two organic semiconductors with mobile anions and cations, respectively. Mobile ionic charge was successfully deployed to control the direction of electronic current flow in semiconductor devices. As a result, these devices showed electroluminescence under forward bias and a photovoltage upon illumination with visible light. Thus, ionic charge carriers can enhance the performance of existing electronic devices, as well as enable new functionalities.**

Junctions between n-type and p-type semiconductors are the cornerstone of modern electronic materials technology because they enable a variety of solid-state devices such as transistors, light-emitting diodes, and photovoltaic cells (*1*). Diffusion of electronic charge across a pn junction sets up a built-in potential, which allows current to flow preferentially in one direction (rectification). Rectification is also observed in junctions of ionic conductors, where a built-in potential arises from diffusion of anions (N) and cations (P) across a membrane. Such ionic junctions play an important role in biology. In bilayer membranes, for example, voltage-gated ion channels control the direction of ion transport into and out of cells (*2*). Although such systems provide an inspiration for rectifying devices, their inherent complexity makes it difficult to study systematically. Electrolytic junctions formed by using positively and negatively charged polymer solutions offer a synthetic alternative. For example, by defining the interface with a permeable membrane, a junction was formed between a solution of a polymeric acid and a solution of a polymeric base, with mobile protons ($H^+$) and hydroxide ions ($OH^-$), respectively. Under an applied alternating current, this type of junction showed considerable rectification of ionic current (*3*).

The analogy between electronic (pn) and ionic (PN) junctions has not been exploited in solid-state devices. In addition to the fundamental merit of exploring the coupling between ionic and electronic carriers, such junctions may result in devices with improved performance and new functionalities. Progress toward this goal has been hampered by a lack of materials that combine semiconducting behavior with appreciable ionic conductivity. The

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addition of salts in films of conjugated polymers led to devices in which ion concentration gradients enhance the injection of electronic charge and lead to efficient electroluminescence (*4*). These gradients in ion concentration are established by an applied bias. As a result, these devices do not show inherent rectification, and the ion gradients tend to disappear when the bias is removed (*4*). The electrochemical deposition of junctions of polyacetylene films with oppositely charged ionomers has led to rectification (*5*). However, the rectification was attributed to an asymmetry in the ion polarization processes near the metal electrodes and the inability of one of the layers to transport electrons. Removal of the mobile ions led to the formation of purely electronic pn junctions (*6*). Electroluminescence or photovoltaic response was not reported in these devices. Electrochemical deposition also does not produce abrupt junctions and is of limited applicability to a subset of organic materials.

Over the past few years, several organic semiconductors with intrinsic ionic conductivity have emerged. A particular example is ionic transition metal complexes, such as $[Ru(bpy)_3]^{2+}(PF_6^-)_2$, where bpy is 2,2′-bipyridine (Fig. 1). These materials, studied extensively in electrochemistry and spectroscopy (*7*), have recently attracted renewed interest in solid-state electroluminescent devices (*8*–*10*). The $[Ru(bpy)_3]^{2+}$ ion is an intrinsic semiconductor with a negligible concentration of mobile electrons and holes at room temperature due to a large HOMO-LUMO (highest occupied molecular orbital–lowest unoccupied molecular orbital) gap (*7*). However, substantial concentrations of electrons and holes can be injected into a film from metal electrodes where they migrate by hopping and recombine to give rise to light emission (*11*). An identifying feature of these materials is the mobile counter ions ($PF_6^-$), which redistribute under the application of an applied bias and assist the injection of electronic charge (*11*). The redistribution of counter ions is usually slow, giving rise to a characteristic delay between the application of a bias and the emission of light (*12*).

In addition to ionic transition metal complexes, any organic semiconductor can, in principle, be modified with the addition of appropriate groups that endow ionic conductivity. Considerable interest has focused on conjugated polymers with ionomers attached on side chains (*13*). Small molecules can be similarly modified. One example is the molecule 9,10-diphenylanthracene-2-sulfonate with sodium counter ions ($DPAS^-Na^+$) (Fig. 1), which has been used as a water-soluble fluorescence probe and for the generation of electrogenerated chemiluminescence in solution (*14*, *15*). The ability to introduce ionic conductivity to practically any organic semiconductor enables the synthesis of materials with a broad range of electronic and ionic transport characteristics.

In analogy to semiconductors, ionic conductivity can be classified as N-type in materials such as $[Ru(bpy)_3]^{2+}(PF_6^-)_2$, where the anions are the predominant carriers of ionic current, and P-type in materials such as $DPAS^-Na^+$, where the opposite is true. Because both N-type and P-type ionic conductors are available, ionic (PN) junctions can be fabricated in direct anal-

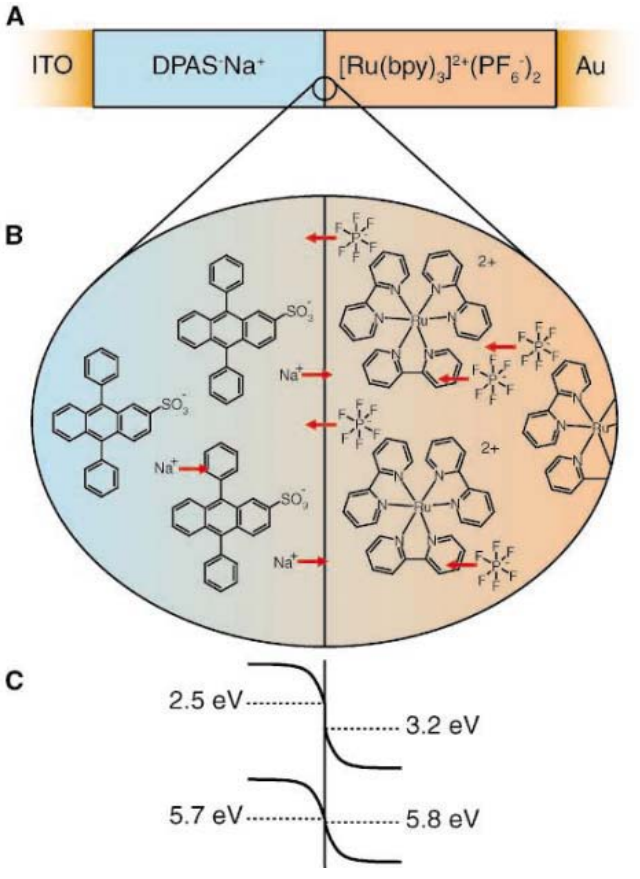


**Fig. 1.** Structure and energetics of the PN junction. (**A**) Structure of the PN junction. (**B**) Detail of the junction area. (**C**) Relevant energy levels at the junction before (dotted lines) and after (solid lines) ion diffusion. The arrows in (B) indicate the direction in which the counter ions diffuse to form the junction when the two layers are brought in contact.

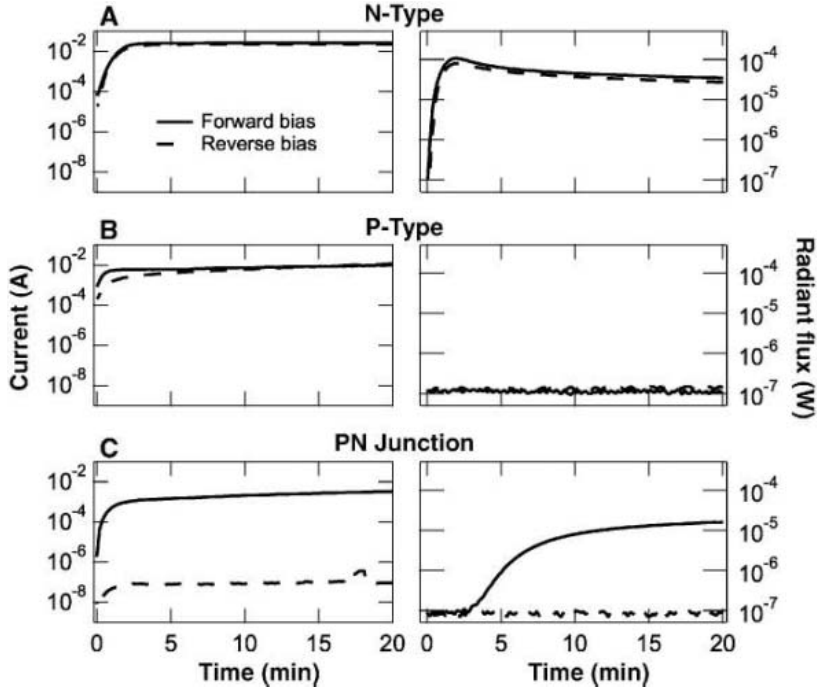


**Fig. 2.** Temporal response of the current and radiance of the various devices at forward and reverse bias. (**A**) ITO/$[Ru(bpy)_3]^{2+}(PF_6^-)_2$/Au, (**B**) ITO/$DPAS^-Na^+$/Au, and (**C**) ITO/$DPAS^-Na^+$//$[Ru(bpy)_3]^{2+}(PF_6^-)_2$/Au.

ogy to conventional semiconductor electronic (pn) junctions (*1*). Mechanistically, such a PN junction is similar to a pn junction: Gradients in the concentration of mobile ions at the junction result in diffusion of cations into the N-type material and anions into the P-type. As ions diffuse, a built-in potential that opposes diffusion is established. Equilibrium is reached when the flux due to ion diffusion is balanced by the flux due to ion drift caused by the built-in potential. This built-in potential modifies the energy levels for electrons and holes at the junction, in a manner analogous to band bending in crystalline semiconductors. Similar to a pn junction, the built-in potential directs photogenerated electronic carriers toward opposite electrodes. The interplay between junction energetics (determined by the semiconductor molecules) and built-in potential (induced by the redistribution of ions) provides a powerful means of controlling device performance.

The fabrication of such junctions is challenging. Vapor deposition techniques, which are usually used to fabricate high-quality heterojunctions of organic semiconductors, are of limited use due to the low vapor pressure of ionic materials. Successive deposition from solution is also challenging because dissolution of the bottom layer leads to intermixing of the two materials, generally precluding the formation of an abrupt junction. These problems were circumvented by using the technique of soft-contact lamination (*16*, *17*). A $DPAS^-Na^+$ film was deposited on a glass substrate with patterned indium tin oxide (ITO) electrodes. A $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ film was deposited on a polydimethylsiloxane (PDMS) substrate with patterned Au electrodes. The two substrates were brought together, laminating the two organic layers to yield the structure ITO/$DPAS^-Na^+$//$[Ru(bpy)_3]^{2+}(PF_6^-)_2$/Au (Fig. 1), where // indicates the location of the laminated interface. The elastomeric PDMS substrate enabled the lamination by conforming to the contours of the bottom half of the device, and the transparent ITO electrode allowed optical access to the junction (*18*).

To better understand the operation of the PN junction, it is important to consider the behavior of its components. The temporal evolution of the current and radiant flux of ITO/$[Ru(bpy)_3]^{2+}(PF_6^-)_2$/Au devices at ±3V are shown in Fig. 2A. The curves show the usual response for organic semiconductor devices with mobile ions, with a slow turn-on time consistent with redistribution of the $PF_6^-$ counter ions (*11*). At steady state, the device does not show rectification, consistent with previous reports (*11*). The lack of rectification is a result of the ionic nature of this material. When ITO is biased positive with respect to Au, the $PF_6^-$ counter ions accumulate near the ITO electrode, leaving uncompensated $[Ru(bpy)_3]^{2+}$ near the Au electrode. The ionic charge leads to high electric fields near the electrodes, which help inject holes from ITO and electrons from Au into the $[Ru(bpy)_3]^{2+}$ molecules. Reversing the bias leads to accumulation of the $PF_6^-$ counter ions near the Au electrode, and the electric fields at the interface assist the injection of electrons from ITO and holes from Au (*11*). Similar characteristics were obtained for the P-type layer (Fig. 2B). The emission from the ITO/$DPAS^-Na^+$/Au device was below the noise threshold of the integrating sphere/photodetector assembly. It was estimated in a separate experiment to be on the order of $10^{-11}$ and $10^{-10}$ W for reverse and forward bias, respectively. These results show that both electrons and holes are injected and are mobile in $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ and $DPAS^-Na^+$ films.

In contrast to its constituent layers, the PN junction shows pronounced rectification. Figure 2C shows the current in ITO/$DPAS^-Na^+$//$[Ru(bpy)_3]^{2+}(PF_6^-)_2$/Au to be more than $10^4$ times higher under forward bias (ITO positive) than under reverse bias (±5 V, 20 min after application of a bias). At the junction, counter ion diffusion is expected to lead to excess negative ionic charge in the P-side of the junction and excess positive ionic charge in the N-side of the junction. The resulting built-in potential raises the HOMO and LUMO levels of $DPAS^-$ relative to $[Ru(bpy)_3]^{2+}$ (Fig. 1C), which suppresses the transport of holes from $DPAS^-$ to $[Ru(bpy)_3]^{2+}$ (and the transport of electrons in the opposite direction). However, at short-circuit (zero applied bias), the Fermi levels of the two metals will align. This will be accommodated by additional potential drops near the metal contacts, caused by ion redistribution (*18*). These potential drops assist the injection of holes from ITO in $DPAS^-$ and electrons from Au in $[Ru(bpy)_3]^{2+}$. Under an applied bias, the steady-state current is determined by the complex interplay between ion-induced potentials, the potentials associated with injected electronic carriers, and the energy levels of the constituent materials. Device simulations (fig. S2) show that the magnitude and the spatial extent of the built-in potential vary with bias as in a pn junction. To the first order, the application of forward bias suppresses the built-in potential and allows holes from $DPAS^-$ to be injected in $[Ru(bpy)_3]^{2+}$ (and vice versa for electrons). At the same time, enhanced charge injection from the metal contacts takes place. Reverse bias increases the magnitude of the built-in potential at the junction. As a result, only a small fraction of the applied bias drops near the contacts to help inject electronic carriers, leading to poorer injection and an overall lower current than under forward bias. As expected, the direction of current flow was reversed in control devices in which the order of the two layers was reversed (ITO/$[Ru(bpy)_3]^{2+}(PF_6^-)_2$//$DPAS^-Na^+$/Au).

The PN junctions show intense light emission under forward bias, with a luminance of

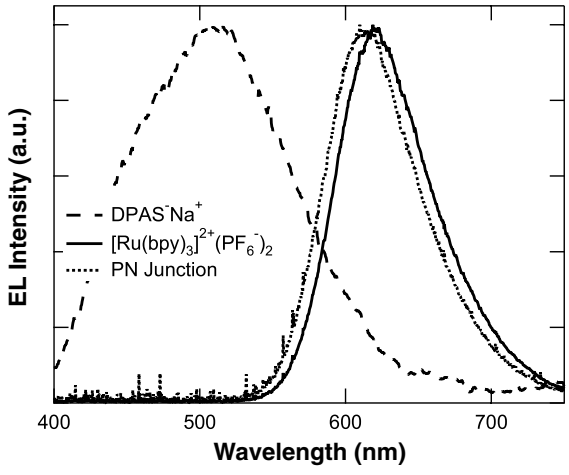


**Fig. 3.** Electroluminescence spectra of the various devices.

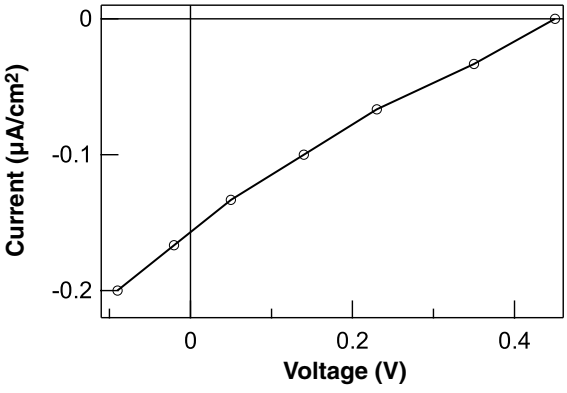


**Fig. 4.** Photovoltaic response of the PN junction.

500 cd/m$^2$ at 5 V. Their spectral response (Fig. 3) reveals that emission arises from the $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ side of the junction. This is consistent with the energy-level offsets at the heterojunction (Fig. 1C). The barrier for hole injection from $DPAS^-Na^+$ into $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ is 0.7 eV smaller than that for electron injection from $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ into $DPAS^-Na^+$. No light emission was detected at reverse bias (the radiant flux was lower than $10^{-12}$ W). In the absence of mobile ions, a device with energy levels as in Fig. 1C (dotted lines) and with ITO and Au electrodes would show hole-only current without substantial rectification or light emission. Rectification and light emission in traditional organic light-emitting diodes are associated with the use of an anode and a cathode with a high and a low work function, respectively.

Illumination of the junctions resulted in a photovoltaic response. Under 100 mW/cm$^2$ excitation from a halogen lamp, an open circuit voltage of 0.45 V and a short-circuit current density of 0.15 μA/cm$^2$ were measured (Fig. 4). Upon sudden illumination the photocurrent took a few seconds to reach steady state, presumably due to redistribution of the ionic carriers under the influence of the photogenerated electrons and holes. The data of Fig. 4 were acquired at steady state. The intensity dependence of the photocurrent was measured up to 500 mW/cm$^2$ and was found to be sublinear, with an exponent of 0.85. The internal quantum efficiency (electrons collected per photon absorbed) was estimated to be on the order of $5 \times 10^{-5}$. This is low compared to state-of-the-art organic photovoltaics but expected given the low exciton dissociation yield in these materials. Indeed, devices based on the constituent layers did not show any photovoltaic response, confirming negligible exciton dissociation in the bulk and at the electrode interfaces.

The photovoltaic response of the junctions is consistent with the built-in potential assisting the dissociation of excitons and the separation of electrons and holes in the $[Ru(bpy)_3]^{2+}(PF_6^-)_2$ and the $DPAS^-Na^+$ layers, respectively (Fig. 1C). Estimating the magnitude of the built-in potential is difficult, because the ion density near the junction interface is hard to predict due to steric effects associated with ion packing. The value of the open circuit voltage represents a lower limit for the built-in potential, considering that there is some resistive loss across the two layers and at the contacts. It should be mentioned that the use of mobile ions in photovoltaics is well established in Grätzel cells (*19*). In these devices, ions in an electrolyte extract and transport holes from charge-transfer dyes attached to titanium dioxide. In the ionic junctions reported here, the ions establish a built-in potential across a heterojunction.

The observations of rectification in current and light emission, and of a photovoltaic response in the PN junction and not in the constituent layers, demonstrate that mobile ionic charge can be used to control the flow of electronic current in solid-state devices. In principle, any organic semiconductor can be modified with ionomers to endow unipolar ionic conductivity, and the technique of lamination described here can be easily applied to fabricate PN junctions from these materials. Such junctions might help decrease recombination and increase the efficiency of organic heterojunction solar cells (*20*). Moreover, given that ionic and electronic mobilities often differ by several orders of magnitude, the possibility of realizing ionic junctions that can be reconfigured by the prolonged application of a bias and be used at faster time scales to rectify electronic current is exciting.

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# Tectonic Uplift and Eastern Africa Aridification


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**The history of Eastern African hominids has been linked to a progressive increase of open grassland during the past 8 million years. This trend was explained by global climatic processes, which do not account for the massive uplift of eastern African topography that occurred during this period. Atmosphere and biosphere simulations quantify the role played by these tectonic events. The reduced topographic barrier before 8 million years ago permitted a zonal circulation with associated moisture transport and strong precipitation. Our results suggest that the uplift itself led to a drastic reorganization of atmospheric circulation, engendering the strong aridification and paleoenvironmental changes suggested by the data.**


Several biosphere shifts linked to phases of aridification punctuated East African environmental evolution during the late Neogene (*1*, *2*) 8 million to 2 million years ago (Ma), which corresponds to a key period in hominid evolution (*3*–*5*). Isotopic studies (*1*) have attributed a first Upper Miocene (8 to 6 Ma) transition from woodlands to grasslands to atmospheric $CO_2$ decrease. A later (5 to 3 Ma) spreading of grasslands (*6*, *7*) was attributed to both Indian Ocean sea surface temperature cooling (*8*) and the onset of glacial-interglacial cycles (*9*), whereas topographic changes induced by rifting processes were considered as a second-order forcing factor on climate.

Actually, substantial uplifts affected African topography during the late Neogene. The most important topographic structure is the East African Rift System. It began to uplift by its eastern branch (in southern Ethiopia and the Turkana depression in northern Kenya) during Eocene-Oligocene times, with uplifting reaching a maximum at the Plio-Pleistocene interval (*10*–*12*) (Fig. 1D). Ethiopian uplift shoulders were superimposed on an older topography linked to Oligocene volcanic activity (Ethiopian traps) (*13*). Traps sequence thickness is not very well constrained—a maximum of 2000 m having been observed on the northwestern edge

of the Ethiopian plateau (*13*). The western branch of the East African Rift System started to develop during the middle-late Miocene, with initiation of the central Tanganyika Basin at about 12 to 10 Ma (*14*, *15*), and with more recent phases of major uplift between 5 and 2 Ma in the Tanganyika and Malawi rifts (*16*). Major Tanzanian escarpments were present by 3 Ma (*17*). These features led to a 6000-km-long elevated area (*11*), mostly oriented north-south and bordered by crests culminating between 1500 and 5100 m. The Karoo plateau in South Africa (20° to 32°S) has been raised during the past 5 million years (*10*), reaching a mean elevation of ~1400 m at present. Modeling studies of the impact of Cenozoic uplift on climate have been essentially carried out at the global scale, and Mio-Pliocene orographic changes have been implicated as a link between Tibetan plateau uplift and monsoons (*18*). Nevertheless, the impact of African topography on climate and vegetation has not yet been tested.

Using numerical modeling, we assess both climate and biosphere responses to topographic changes linked to eastern and southern African uplifts. Sensitivity experiments have been carried out with the LMDz4 (Laboratoire de Météorologie Dynamique, Paris) (*19*) atmospheric general circulation model (AGCM). To improve the AGCM's ability to simulate climate change induced by the narrow topographic structures of the East Africa Rift System and the Southern African topography, we used a zoom giving a resolution of up to 1° over our region of interest. In addition to a present-day control (CTL) simulation, we made two runs with a reduced topography over eastern and southern Africa (Fig. 1, A to C). As reconstructions for Eastern Africa paleoaltitudes are not available because of a lack of accurate data, we constrained our model according to two extreme scenarios from geological literature (*20*). The NORIFT simulation considers the Ethiopian traps as being very low. Topography is reduced to 400 m over both eastern and southern Africa. In the TRAPS simulation, we account for a maximum elevation of Ethiopian traps at 2000 m (*13*), and we set this value as minimal topography over the traps area. Both simulations were prescribed with present-day boundary conditions, namely modern sea surface temperatures, ice-sheet and sea-ice extent, insolation, and greenhouse gas concentrations. Climatic outputs have been used to force the dynamic global vegetation model Organizing Carbon and Hydrology in Dynamic Ecosystems (ORCHIDEE) (*21*). Climatic variables (*20*) have been derived from our three simulations to force the biosphere model. Here, we consider the potential vegetation cover with ORCHIDEE plant functional types (PFTs).

CTL rainfall patterns are in good agreement with reanalysis data (*22*), except at the land-sea interface of the East African coast where the precipitation amount is overestimated. Present-

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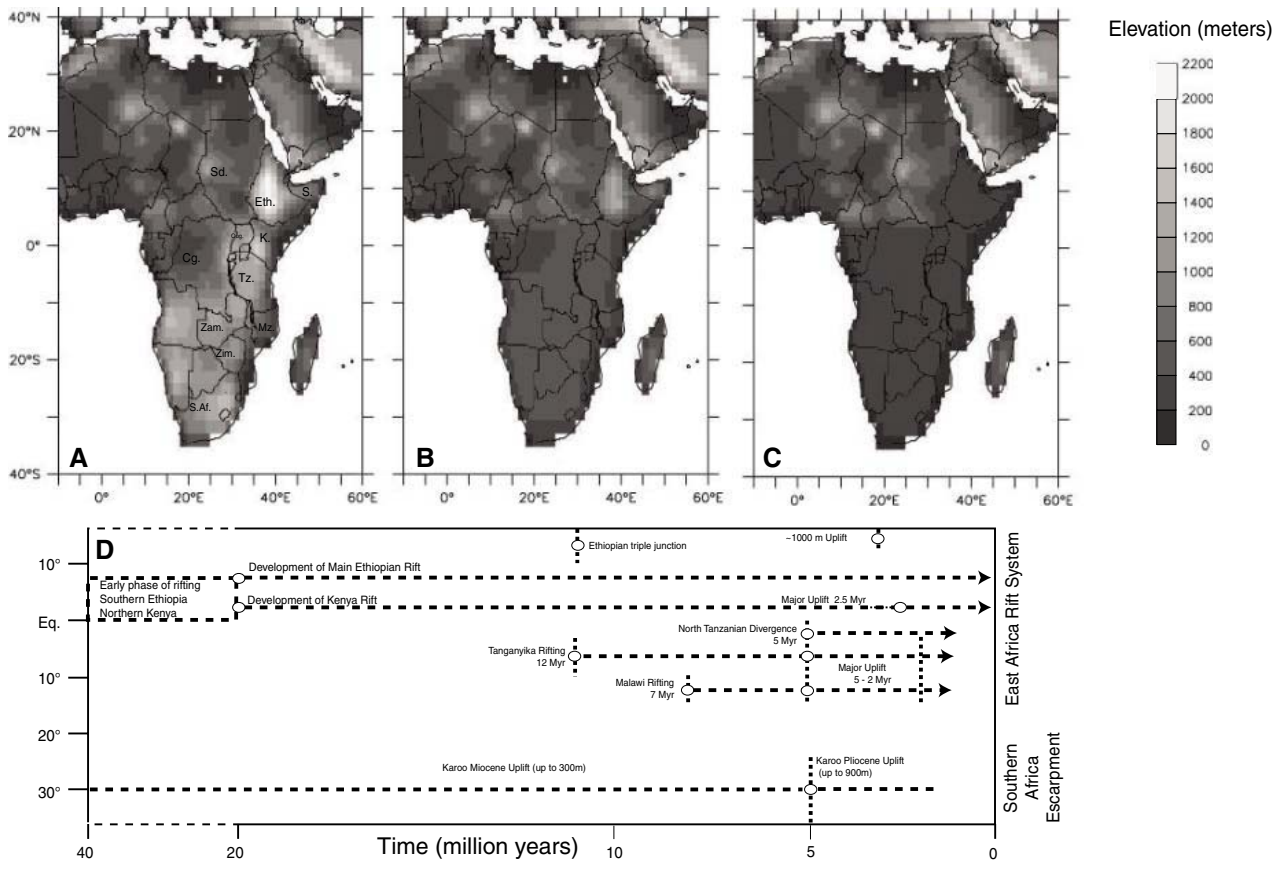


**Fig. 1.** Experimental design of the three LMDz4 simulations related to topography history of Africa. (**A** to **C**) Relief interpolated at the AGCM resolution for CTL, TRAPS, and NORIFT runs, respectively. (**D**) Simplified history of major African tectonic events during the late Neogene. We note that several phases of uplift occurred during the last 10 million years, a period during which numerous environmental shifts are recorded in East Africa. Tectonic and paleotopographic data as follows: Ethiopia (*11*–*13*), Kenya (*11*, *12*), Tanzania (*18*), Tanganyika (*15*, *16*), Malawi (*17*), and southern Africa (*11*). Ovals indicate onset of the most important tectonic movements.

day simulated annual rainfall averages 1310 mm over the horn of Africa [0° to 15°N/35° to 50°E, the Ethiopia-Kenya-Somalia (EKS) region], ranging from 200 to 800 mm for southwestern Ethiopia and Somalia up to 2400 mm in northern Kenya. In these regions, rainfall is strongly influenced by low tropospheric winds (*23*), which convey a large amount of atmospheric moisture. These winds blow northeasterly during boreal winter and southwesterly during boreal summer. Continental topography strongly influences the direction of these winds and therefore the location of rainfall. Meridional moisture transport shows that Ethiopian highlands deflect the northeast monsoon flow southward along the Somalian coast during winter and that Kenyan highlands deflect the southeast flow northward during summer (Fig. 2, A and B). When the Ethiopian highlands are reduced in height (TRAPS), the Indian monsoon flow enters more deeply westward over the continent during winter, whereas in summer a zonal eastward moisture flow is created from Southern Sudan to Ethiopia (5° to 10°N) (Fig. 2, C and D). This phenomenon, which leads to large-scale rainfall, is enhanced in NORIFT, as no topographic barrier blocks lower atmosphere circulation over Ethiopia (Fig. 2, E and F). Moreover, higher surface temperatures due to lower altitude in our experiments induce more intense convection and associated precipitation. As a consequence, TRAPS rainfall averaged over EKS is increased by 15% (1520 mm/year) with respect to the CTL experiment (Fig. 3, A and B), whereas it is increased by 40% (1830 mm/year) in NORIFT (Fig. 3C). The seasonal pattern is conserved in TRAPS, with rainfall equally increased in winter and summer. However, the total removal of topography favors winter convective rainfall and leads to a peak of more than 4 mm/day in February in NORIFT (Fig. 3E). South to the equator, a winter eastward moisture transport is set between Atlantic and Indian oceans when topography is reduced. Summer westward Indian flow is no longer diverted by Tanzanian relief and penetrates inland between the equator and 25°S. Once again, rainfall is enhanced: Averaging 1150 mm/year over 0° to 20°S/25° to 40°E, the Congo-Tanzania-Zambia-Mozambique (CTZM) region for CTL, it increases by 38% (1590 mm/year) in TRAPS to 62% (1870 mm/year) in NORIFT (Fig. 3D).

Apart from the overestimated vegetation cover over the Tanzanian and Kenyan coasts due to the LMDz4 positive bias in rainfall already mentioned, the CTL "arboreal" and "herbaceous" fractions are in good agreement with preindustrial values, which can be inferred from present-day data and modeling studies (*24*). Total vegetation cover (Fig. 4A) is globally increased over both eastern EKS and CTZM regions in the TRAPS and NORIFT simulations (Fig. 4, B and C). When the maximal Ethiopian traps elevation is conserved (Fig. 4, B to E), arboreal vegetation is slightly increased over eastern Ethiopia. It becomes dominant over almost all the northeastern rift when topography is totally eliminated (Fig. 4F). Between 15°S and 5°N, the positive shift in arboreal group is the highest, with a transition from 10% to 60% coverage over Ethiopia, Kenya, and Mozambique (NORIFT). These parts of eastern Africa become tree-dominated, whereas they are dominated by herbaceous groups in the present-day experiment (Fig. 4D). In both lowered-topography experiments, we also simulated an arboreal increase connecting western and eastern Africa along the equator. These results suggest a strong impact of Mio-Pliocene uplift on biosphere, and available data depict strong paleoenvironmental changes during this time interval.

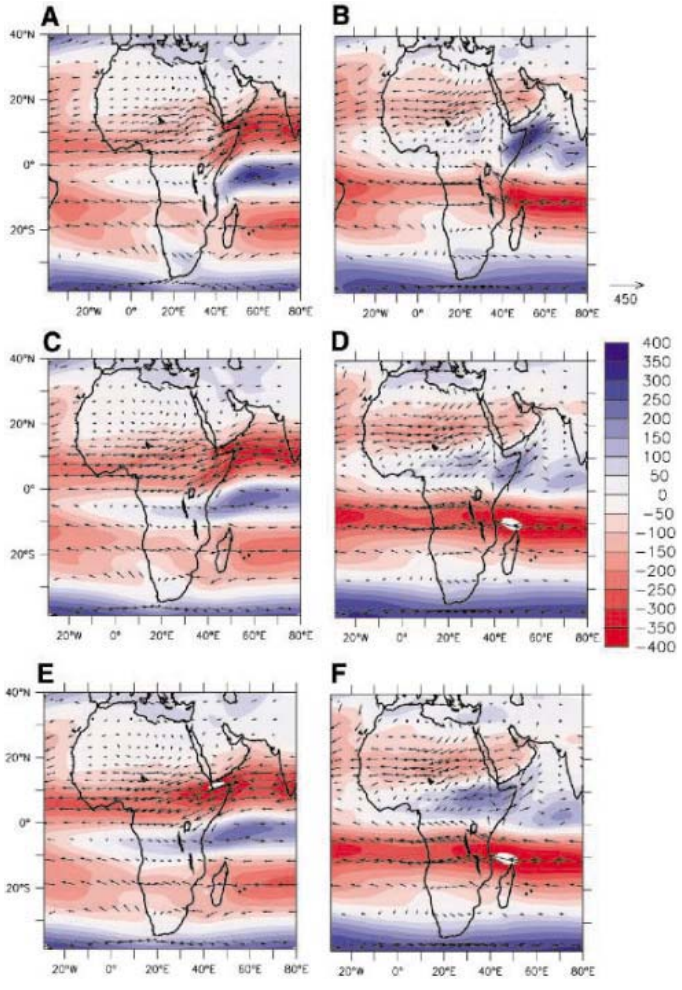


**Fig. 2.** Vertically integrated zonal moisture transport (kg $m^{-1}$ $s^{-1}$) for the three experiments: Winter (**A**, **C**, and **E**) and summer (**B**, **D**, and **F**) transports for CTL, TRAPS, and NORIFT, respectively. The arrow indicates scale. Red and blue indicate westward and eastward moisture transport, respectively. We note the intensification of zonal transport both in winter and summer when topography is reduced.

Between 8 and 6 Ma, new families of ungulate mammals with a $C_4$ (grass-dominated) diet replaced more archaic middle Miocene communities (*25*) with $C_3$ (wood-dominated) diets, indicating a decrease of rain forests and an increase of more open "savannah-mosaic" habitats. Isotopic studies on fossil tooth enamel (*1*) have suggested a global-scale replacement of $C_3$-photosynthesizing plants by $C_4$ plants. This change has been linked to atmospheric $CO_2$ decrease, although the question is still debated (*26*, *27*). Paleoenvironmental data compiled by Cane and Molnar (*8*) for the past 15 million years suggest a later transition, with wooded habitats existing until 5 and 3.7 Ma in northern and southern Kenya, respectively, and until 3.4 Ma in Ethiopia. Until this time interval, moist environments would have been present in northeastern Africa (*28*), notably in southern and central Ethiopia (*29*). The Turkana Basin experienced a strong increase in the number of mammal species adapted to open grasslands, indicating increased aridity after 2.5 Ma (*7*). This trend to aridification at around 3 Ma was first explained by Indian Ocean cooling associated with a change in Indonesian throughflow conformation, which allowed colder water to come in from the Pacific ocean (*8*). Moreover, paleoclimatic records (*30*) suggest an orbital-scale variability in northeast Africa paleoenvironments that increased with the intensification of high-latitude glacial cycles (*9*). The topographic impact has been considered as a second-order forcing so far, only modulating the variability of regional climate through rain-shadow effects (*2*).

Despite the high resolution we used, our experiments cannot capture isolated topographic

features like volcanoes or small mountains. Nevertheless, our results provide strong arguments for an impact of topographic changes at the continental scale. Topography clearly affects moisture transport and, therefore, rainfall spatial patterns and amounts. Associated hydrological modifications induce strong shifts in vegetation. Consequently, it appears that uplifts had a first-order impact on Mio-Pliocene eastern African climate evolution. We cannot pretend to have captured an exhaustive picture of vegetation evolution in the several rift basins of eastern Africa, but these results are a step forward in understanding the mechanisms that led to more open landscapes in eastern Africa. We can assume that the climatic consequences depicted here, along with landscape fragmentation linked to asynchronous uplift events, have largely contributed to the setting of present-day vegetation patchwork of eastern and southern Africa. Consequently, African uplift must be considered as a dominant forcing of the late Neogene climate of eastern Africa, and not as a background factor. Future studies, which will aim to accurately assess the impact of climate change at this period, and notably its potential influence on paleoenvironments that force hominid evolution, will have to constrain accurately the topographic history of eastern Africa.

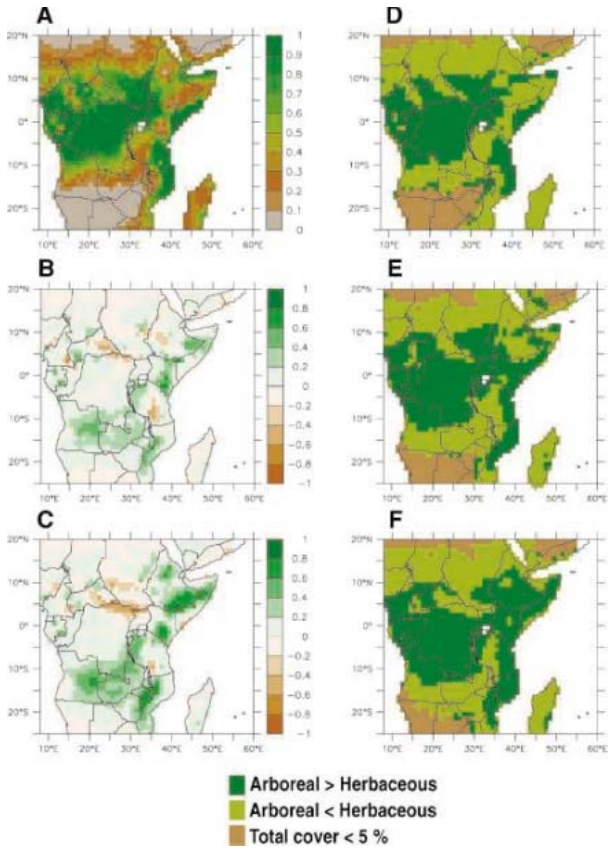


**Fig. 4.** Simulated vegetation for the three experiments. (**A**) Summed vegetation fraction over every PFT for the CTL run. Zero value indicates the absence of vegetation; 1 represents a gridpoint totally covered by PFTs. (**B** and **C**) Anomalies of TRAPS and NORIFT minus CTL, respectively. (**D** to **F**) Arboreal-dominant, herbaceous-dominant, and desert-like fractions for CTL, TRAPS, and NORIFT, respectively. We note the massive spreading of arboreal fraction at the expense of herbaceous fraction over eastern Africa.

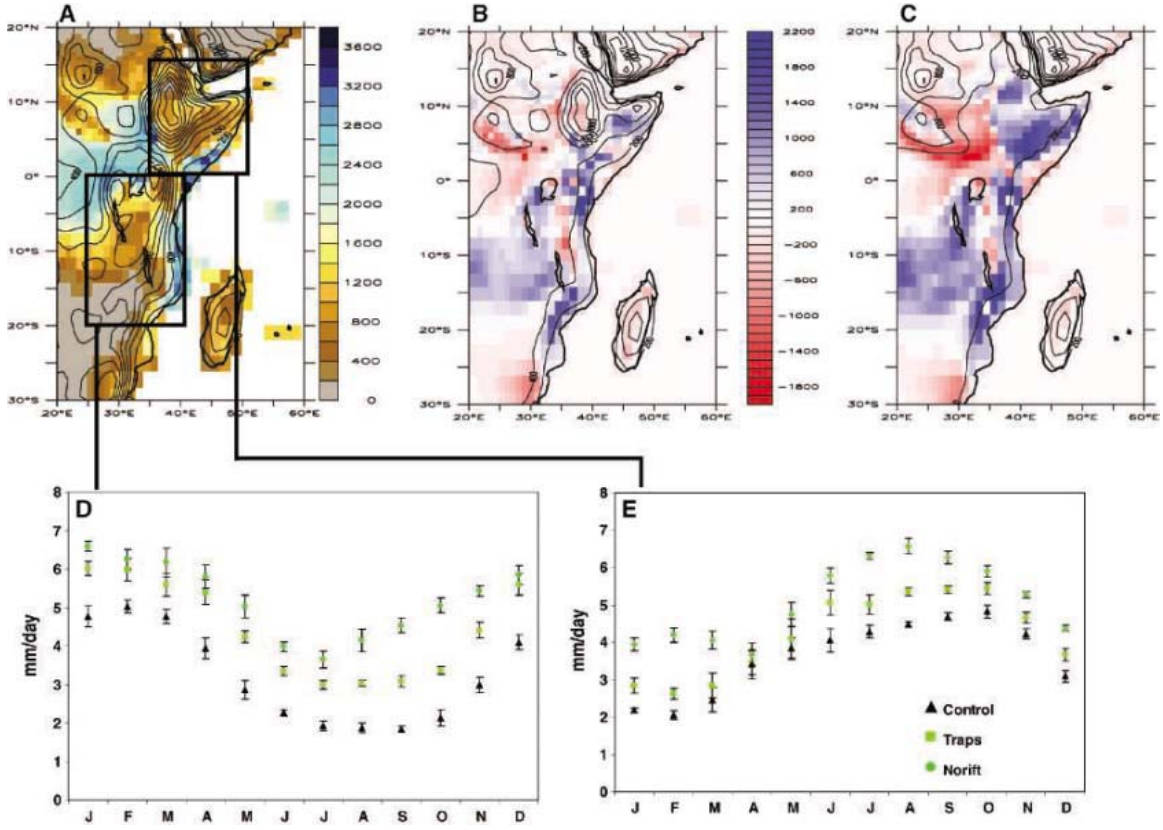


**Fig. 3.** Simulated rainfall for the three experiments. (**A**) Annual amount of rainfall simulated by CTL run. (**B** and **C**) Rainfall anomaly of TRAPS and NORIFT runs versus CTL, respectively. Isolines represent topography for each run, in meters. (**D** and **E**) Seasonal pattern of rainfall averaged over EKS and CTZM (black boxes), respectively. Rainfall variability is calculated excluding the unbalanced first year of simulation.

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# Measurement of the Entanglement of Two Superconducting Qubits via State Tomography


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**Demonstration of quantum entanglement, a key resource in quantum computation arising from a nonclassical correlation of states, requires complete measurement of all states in varying bases. By using simultaneous measurement and state tomography, we demonstrated entanglement between two solid-state qubits. Single qubit operations and capacitive coupling between two superconducting phase qubits were used to generate a Bell-type state. Full two-qubit tomography yielded a density matrix showing an entangled state with fidelity up to 87%. Our results demonstrate a high degree of unitary control of the system, indicating that larger implementations are within reach.**


The laws of quantum physics provide intriguing possibilities for a tremendous increase in computational power compared with classical computation (*1*). Because this power is achieved through the controlled evolution of entangled quantum states, a clear demonstration of entanglement represents a necessary step toward the construction of a scalable quantum computer (*2*, *3*). However, direct demonstration of entanglement is challenging because all of the DiVincenzo criteria (*4*) for quantum computation must be met simultaneously. To date, only subsets of these key requirements have been demonstrated for superconducting qubits (*5*–*9*). We demonstrated all of the DiVincenzo criteria simultaneously, thus placing superconducting qubits on the road map for scalable quantum computing.


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Circuits made of superconductors and Josephson junctions are promising candidates for scalable quantum computation because of their compatibility with integrated-circuit fabrication technology (*5*–*9*). The Josephson phase qubit stands apart from other superconducting qubits because it does not use an optimal operating point. Coupling of phase qubits is thus straightforward, allowing for multiple control methods (*10*). With recent improvements in coherence times and amplitudes (*11*), and in particular the ability to measure both qubit states simultaneously (*5*), it is possible to use phase qubits to produce entangled states and measure them with high fidelity.

In the phase qubit circuit (Fig. 1A), the Josephson junction (with critical current $I_0$) has a superconducting phase difference, $\delta$, that serves as the quantum variable. When biased close to the critical current, the junction and its loop inductance, $L$, give a cubic potential that has qubit states $|0\rangle$ and $|1\rangle$, with an energy spacing that corresponds to a transition frequency $\omega_{10}/2\pi \sim 5$ GHz (Fig. 1B). This frequency can be adjusted by ~30% via the bias current.

Single qubit logic operations, corresponding to rotations about the $x$, $y$, and $z$ axes of the Bloch sphere, were generated as follows. Rotations about the $z$ axis were produced from current pulses on the qubit bias line that adiabatically change the qubit frequency, leading to phase accumulation between the $|0\rangle$ and $|1\rangle$ states of the qubit (*11*). Rotations about any axis in the $xy$ plane were produced by microwave pulses resonant with the qubit transition frequency. They selectively address only the qubit energy levels, because transitions to higher-lying energy levels are off-resonance due to the anharmonicity of the potential and the shaping of the pulses (*12*). The phase of the microwave pulses defines the rotation axis in the $xy$ plane. The pulse duration and amplitude control the rotation angle.

The qubit state was measured by applying a strong pulse, $I_z$, so that only the $|1\rangle$ state tunnels out of the cubic well (Fig. 1C). Once tunneled, the state quickly decays into an external ground state that can be easily distinguished from the untunneled $|0\rangle$ state by an on-chip superconducting quantum interference device (SQUID) amplifier.

Two separate phase qubits were coupled with a fixed capacitor (*5*) (Fig. 1D). With the qubits labeled A and B, the coupling Hamiltonian is $H_{\mathrm{int}} = (S/2)(|01\rangle\langle 10| + |10\rangle\langle 01|)$, where $|01\rangle = |0\rangle_{\mathrm{A}} \otimes |1\rangle_{\mathrm{B}}$. The coupling strength, $S = (C_x/C)\hbar\omega_{10}$, is proportional to the coupling capacitance $C_x \approx 3$ fF, where $C \approx 1.3$ pF is the junction shunting capacitance (*13*) and $\hbar$ is Planck's constant ($h$) divided by $2\pi$. The two qubits may easily be brought into resonance, even though they are not identical, because each can be tuned over a large frequency range. On resonance, the interaction produces an oscillation with frequency $S/h$ between the states $|01\rangle$ and $i|10\rangle$; for an interaction time of $h/4S$, the coupling produces the gate $\sqrt{i\mathrm{SWAP}}$. This gate, together with single qubit gates, is universal (*14*). The coupling also manifests itself as an avoided level crossing of strength $S/h$ in the spectroscopy of the individual qubits (*15*).

The performance of each qubit can be determined separately by strongly detuning the two qubits relative to $S/h$ so that they behave independently. A standard set of experiments, including Rabi and inversion recovery experiments, gives an energy relaxation time of $T_1$ = 130 ns and a dephasing time of $T_2^*$ = 80 ns for each qubit. These results are consistent with measured values of an uncoupled sample (*11*), indicating no additional loss due to the second qubit. The measurement fidelities, defined as the probabilities of correctly identifying states $|0\rangle$ and $|1\rangle$, are $F_0$ = 0.95 and $F_1$ = 0.85, respectively.

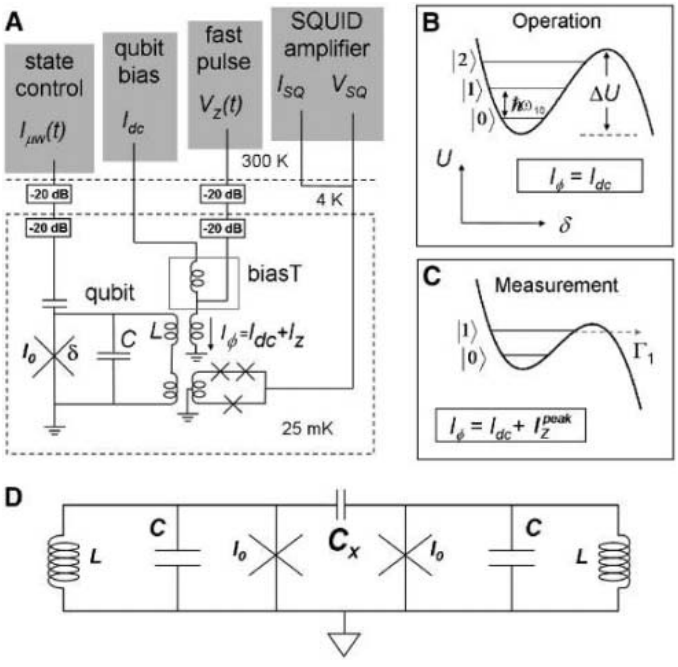


**Fig. 1.** Qubit circuit and experimental operation. (**A**) Circuit schematic for a single Josephson phase qubit, where the X symbol represents the Josephson junction. The measurement is implemented with a broadband 50-ohm transmission line with cold attenuators that is connected to the flux bias line with a bias tee. (**B**) Operation mode of the qubit showing the potential energy, $U$, versus junction phase, δ. The qubit is formed from the two lowest eigenstates $|0\rangle$ and $|1\rangle$, with a transition frequency $\omega_{10}(I_{dc})/2\pi$ = 5.1 GHz that can be adjusted by varying the bias, $I_\phi$. (**C**) Measurement mode of the qubit. During the measurement pulse, the energy barrier $\Delta U$ is lowered to increase the tunneling rate, Γ, and the tunneling probability of $|1\rangle$. (**D**) Circuit diagram of the coupled qubits. The loop inductance, $L$, is ~850 pH, and the junction capacitance, $C$, is ~1.3 pF. An interdigitated capacitor with $C_x \sim 3$ fF couples the qubits, giving rise to an interaction strength of magnitude $S/h$ = 10 MHz.

Our next tuned both qubits to $\omega_{10}/2\pi$ = 5.1 GHz and determined the splitting $S/h$ = 10 MHz by qubit spectroscopy. The time dynamics of the coupling was verified by initializing the qubits to the state $|00\rangle$ and applying to qubit B a 180° rotation about the $x$ axis ($180_x$ pulse) of 10-ns duration. This pulse is sufficiently long to avoid unwanted transitions to other energy levels but short on the time scale of the coupling. The resulting state $|01\rangle$ is not an eigenstate of the coupling Hamiltonian and thus evolves in time according to $|\psi(t)\rangle = \cos(St/2\hbar)|01\rangle - i\sin(St/2\hbar)|10\rangle$. After a variable free-evolution time, $t_{free}$, we simultaneously measure the state of the two qubits. Repeating the experiment about 1000 times, we determine the occupation probabilities $P_{00}$, $P_{01}$, $P_{10}$, and $P_{11}$. This sequence of operations is depicted in Fig. 2A, and the measured probabilities are plotted in Fig. 2B.

The occupation probabilities $P_{01}$ and $P_{10}$ oscillate out of phase with a period of 100 ns, consistent with the spectroscopic measurements. The amplitude and decay of the data are also compatible with the separately measured lifetimes and measurement fidelities of the single qubits. Compared with earlier experiments (*5*), the amplitude of the measured oscillations is substantially larger because of improvements in single qubit fidelities. We note that the oscillations persist longer than the dephasing time, $T_2^*$ = 80 ns, because the period of the coupled qubit oscillations (Fig. 2) is, to first order, insensitive to the detuning of the qubits. For these states, this represents a degeneracy point that is also tunable.

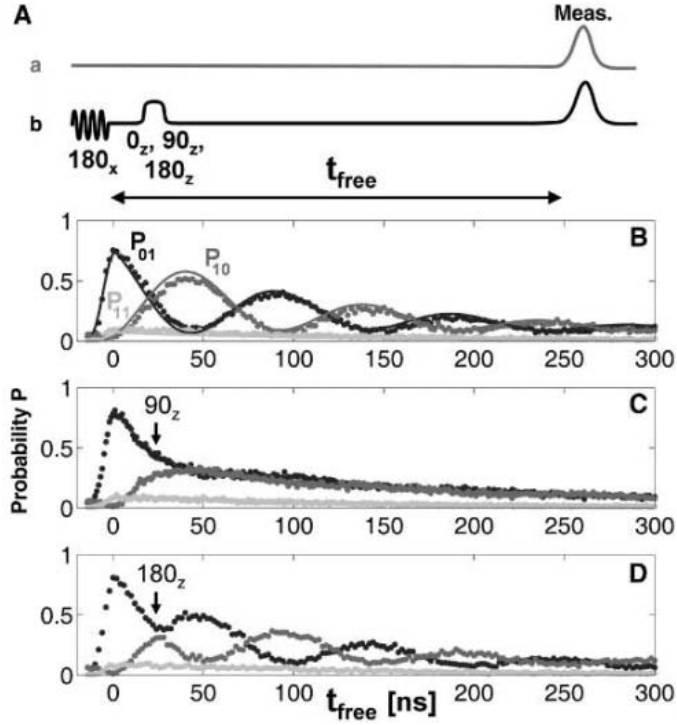


**Fig. 2.** Coherent operations on coupled phase qubits. (**A**) Sequence of operations. A 10-ns-long $180_x$ pulse is applied to qubit B, populating the $|01\rangle$ state. After a free evolution time $t_{free}$ in which the qubits interact, the state occupation probabilities are measured by using 10-ns current pulses that induce selective tunneling of the $|1\rangle$ state. For data in (C) and (D), a $90_z$ and $180_z$ pulse, respectively, is applied to qubit B after 16 ns. (**B**) Plot of measurement probabilities of the states $|01\rangle$, $|10\rangle$, and $|11\rangle$ as a function of $t_{free}$. Note that $P_{00} = 1 - P_{01} - P_{10} - P_{11}$. The solid lines are the results of simulations using known measurement fidelities, relaxation times, and microwave cross talk. (**C**) Plot of measurement probabilities for a sequence that creates the eigenstate $|\psi_2\rangle = (|01\rangle - |10\rangle)/\sqrt{2}$ of the coupling Hamiltonian. After the eigenstate is formed by the $90_z$ pulse, it ceases to evolve with time. (**D**) As in (C), but with an $180_z$ pulse. Here, the phase of the oscillation changes by 180 degrees.

Although these data are consistent with the production of an entangled state at $t_{free}$ = 25 ns, a more stringent test includes performing coherent single qubit operations on this entangled state to verify the predicted unitary evolution of the system. After the application of a $180_x$ pulse on qubit B and a $t_{free}$ of 25 ns, the system is in the entangled state $|\psi_1\rangle = (|01\rangle - i|10\rangle)/\sqrt{2}$. By then applying a $90_z$ pulse on qubit B, we create the Bell state $|\psi_2\rangle = (|01\rangle - |10\rangle)/\sqrt{2}$. Because $|\psi_2\rangle$ is an eigenstate of the coupling Hamiltonian, it should not evolve with time. Implementation of this sequence of operations is complicated by the coupling interaction that occurs during the single qubit operations. Compared with the coupling interaction time, $t_{free}$ = 25 ns, the duration of the single qubit gates $180_x$ and $90_z$ are 10 ns and 4 ns, respectively, and are thus not negligible. The excess coupled interaction during the single qubit gates can be significantly compensated by reducing the free evolution time (*16*) to $t_{free}$ = 16 ns, which we checked numerically. Upon executing this sequence of operations, we verify

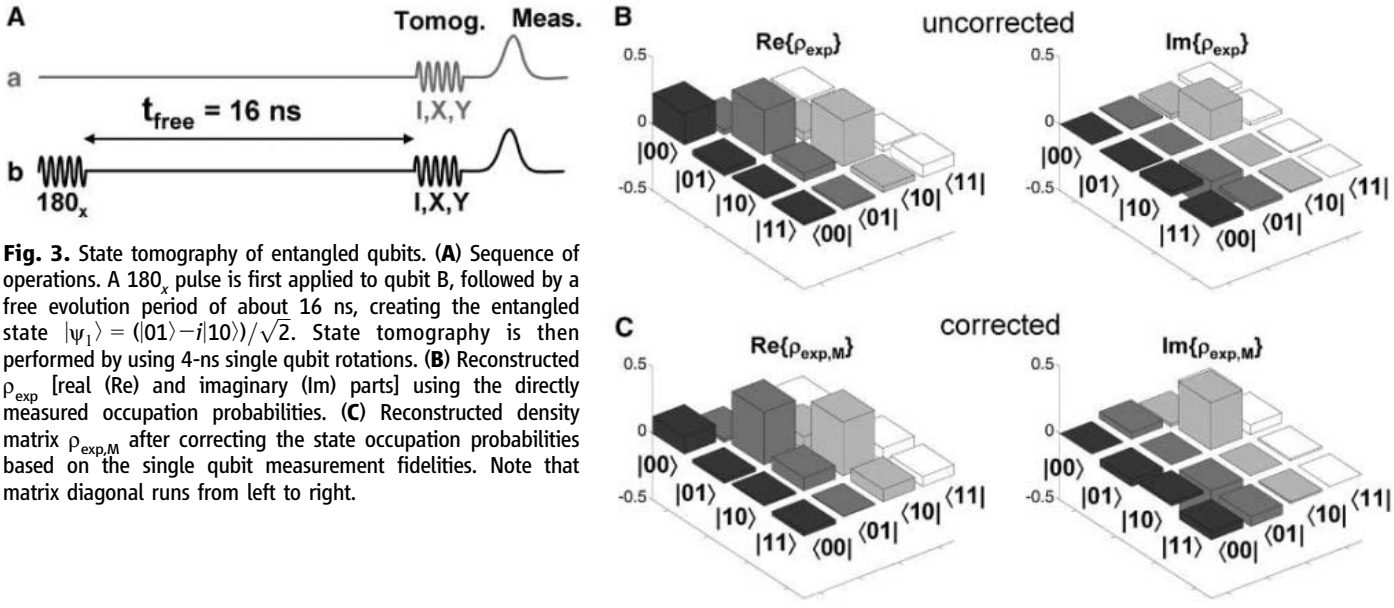


**Fig. 3.** State tomography of entangled qubits. (**A**) Sequence of operations. A $180_x$ pulse is first applied to qubit B, followed by a free evolution period of about 16 ns, creating the entangled state $|\psi_1\rangle = (|01\rangle - i|10\rangle)/\sqrt{2}$. State tomography is then performed by using 4-ns single qubit rotations. (**B**) Reconstructed $\rho_{exp}$ [real (Re) and imaginary (Im) parts] using the directly measured occupation probabilities. (**C**) Reconstructed density matrix $\rho_{exp,M}$ after correcting the state occupation probabilities based on the single qubit measurement fidelities. Note that matrix diagonal runs from left to right.

that indeed $P_{01}$ and $P_{10}$ no longer oscillate as a function of $t_{free}$ (Fig. 2C).

This observed behavior, however, could also be attributed to the destruction of coherence between the states $|01\rangle$ and $|10\rangle$ caused by the application of the $90_z$ pulse. To check this possibility, we applied a $180_z$ pulse on qubit B when the system is in the state $|\psi_1\rangle$, creating the state $|\psi_3\rangle = (|01\rangle + i|10\rangle)/\sqrt{2}$. Because $|\psi_3\rangle$ is equivalent to $|\psi_1\rangle$ but delayed by $t_{free}$ = 50 ns, a reversal of the oscillations is predicted for this experiment. This prediction is verified (Fig. 2D) and provides further evidence of an entangled state.

A full and unambiguous test of entanglement comes from state tomography (*2*, *3*, *17*), which involves the measurement of the quantum state in all nine combinations of three measurement bases ($x$, $-y$, and $-z$) for each qubit. Each measurement gives three unique probabilities (e.g., $P_{01}$, $P_{10}$, and $P_{11}$) for a total of 27 numbers, which are used to compute the 15 independent parameters of the unknown density matrix, $\rho$, via a least squares fit (*17*). The measurement basis change from $-z$ to $x$ and from $-z$ to $-y$ arises from applying a microwave pulse $90_y$ and $90_x$, respectively, before measurement (*11*).

After calibrating the phase of the microwave pulses for the two qubits (*13*), we perform state tomography on $|\psi_1\rangle$ as indicated by the sequence of operations in Fig. 3A. As in the previous experiment, we reduced the duration of the free evolution to compensate for coupled qubit interaction during the initial $180_x$ pulse and the tomography pulses. After executing all nine tomography sequences and measuring the resulting occupation probabilities, we computed the density matrix, $\rho_{exp}$. The real and imaginary parts of the reconstructed $\rho_{exp}$ are shown in Fig. 3B. The imaginary off-diagonal elements $|01\rangle\langle 10|$ and $|10\rangle\langle 01|$ have nearly the same magnitude as the real diagonal components $|01\rangle\langle 01|$ and $|10\rangle\langle 10|$, revealing a coherent superposition of the states $|01\rangle$ and $|10\rangle$. This measurement unambiguously verifies that the two qubits are indeed entangled. Compared to the ideally expected density matrix, $\sigma = |\psi_1\rangle\langle\psi_1|$, we computed the fidelity of the reconstructed quantum state and find $F_{exp} = tr\sqrt{\sigma^{1/2}\rho_{exp}\sigma^{1/2}} = 0.75$.

To identify the sources of fidelity loss, we first corrected for measurement error. Based on the measurement fidelities discussed earlier, we renormalized the measured occupation probabilities and calculated the intrinsic occupation probabilities (*13*). From this we computed a density matrix corrected for measurement, $\rho_{exp,M}$ (Fig. 3C), that gives an improved fidelity, $F_{exp,M} = tr\sqrt{\sigma^{1/2}\rho_{exp,M}\sigma^{1/2}} = 0.87$. We attribute most of the remaining fidelity loss to single-qubit decoherence. By modeling decoherence effects (*16*) using the measured relaxation times, we obtained an expected $\rho_{th}$ that gives a fidelity $F_{exp} = tr\sqrt{\sigma^{1/2}\rho_{th}\sigma^{1/2}} = 0.89$, which is close to the normalized measured value (*18*). The fact that our error is dominated by decoherence indicates good unitary control of our system and thus suggests that improvements in coherence times will directly translate to enhanced gate fidelities. Dramatic increases in coherence should be possible on the basis of straightforward improvements in the dielectric material of the shunting capacitor (*11*, *19*).

Our experiments on coupled phase qubits have verified by state tomography the creation of an entangled Bell state with 87% fidelity. Given that most of the loss in fidelity can be attributed to decoherence, we believe that more complex implementations are well within reach with only modest improvements in qubit coherence times.

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### Supporting Online Material

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# Volcanism in Response to Plate Flexure

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Volcanism on Earth is known to occur in three tectonic settings: divergent plate boundaries (such as mid-ocean ridges), convergent plate boundaries (such as island arcs), and hot spots. We report volcanism on the 135 million-year-old Pacific Plate not belonging to any of these categories. Small alkalic volcanoes form from small percent melts and originate in the asthenosphere, as implied by their trace element geochemistry and noble gas isotopic compositions. We propose that these small volcanoes erupt along lithospheric fractures in response to plate flexure during subduction. Minor extents of asthenospheric melting and the volcanoes' tectonic alignment and age progression in the direction opposite to that of plate motion provide evidence for the presence of a small percent melt in the asthenosphere.

The northwestern Pacific Plate is currently subducting beneath the Kuril and Japan trenches (*1*). This plate dates to the Early Cretaceous [150 to 120 million years ago (Ma) (*2*)] and is dotted with seamounts and oceanic plateaus, interpreted as having formed during Cretaceous times (*3*). The oceanic lithosphere cools as it ages (*4*) and moves away from the divergent plate boundary. Without the presence of a hot spot, new volcanism is not anticipated. During a dive (10K#56 in Fig. 1B) via the remotely operated vehicle *Kaiko*, the sampling of a young alkali basalt [5.95 ± 0.31 Ma (*5*)] near the Japan Trench was unexpected, because there are no known hot spots in the vicinity and recent volcanism has not been reported. In light of this finding, we decided to conduct extensive surveys of the area with the Japan Agency for Marine-Earth Science and Technology research ships *Kairei* (cruises KR03-07 and KR04-08 in Fig. 1B) and *Yokosuka* (cruise YK05-06), which is equipped with the submersible *Shinkai 6500* (dives 6K#877 to 6K#880 in Fig. 1, B and D). Ocean floor mapping was carried out with the use of the multibeam survey system SeaBeam 2112, in the direction of plate motion (Fig. 1A). Two locations were investigated: site A (near the Japan Trench) and site B (located ~600 km to the southeast) (Fig. 1A). These investigations led to the discovery of volcanoes younger than 1 million years old, as well as the presence of a broad lava field. The age difference between the two sites, obtained from plate motion back-calculations, is estimated to be ~6 million years (*6*). The volcanoes proved to be very small (0.005 to 1 $km^3$ each; exposure above the sea floor was estimated from bathymetric data), and we have given them the name "petit spot" volcanoes to convey both their size and their location far from known ridges and hot spots.

Volcanic cones and ridges in site A are distinct from the normal sea floor in bathymetric and sonar surveys (Fig. 1, B and C). Erosion of sediments by the strong bottom current in the trench (*7*) and local normal faulting has exposed the small volcanoes and ridges. These features are composed of moderately to highly vesicular lavas [~10 to 40 volume percent (volume %)], but lavas recovered along the fault outcrops are generally less vesicular (0 to 20 volume %). Previous $^{40}Ar/^{39}Ar$ dating of a lava sample produced an age of ~6 Ma (*5*), and new $^{40}Ar/^{39}Ar$ ages for additional petit spot volcanoes are 4.23 ± 0.19 and 8.53 ± 0.18 Ma (*8*) (table S1).

Approximately 600 km to the southeast of site A lies site B. This site is much farther from the trench and has not been eroded or faulted. Therefore, only the tops of the volcanic cones (<1 km in diameter) stand above the thick surrounding layer of abyssal sediment. Outcrops of young lava are, however, visible in side-scan sonar images because of their high reflectivity (Fig. 1E). Dredges and three *Shinkai 6500* submersible dives (6K#877, 6K#878, and 6K#879) were carried out at three young volcanoes at site B (Fig. 1, D and E). The sequences and lithologies exposed are typical of a single eruption and include highly vesicular (up to 60 volume %) pillow lava; water-chilled bombs; and hyaloclastite, peperite, and contact-metamorphosed mud (*8*) (fig. S1, A and B). Glassy rock samples have 1- to 4-mm-thick palagonitic rims (fig. S1C); because the growth rate of palagonite in basaltic glass exposed to seawater is ~0.003 to 0.02 mm/year (*9*), the volcanic eruption should have occurred between 0.05 and 1 Ma.

Lavas and bombs from sites A and B contained xenocrysts and xenoliths (composed of basalt, dolerite, gabbro, or spinel peridotite), all of which are typical of the oceanic lithosphere (fig. S1C). The presence of xenoliths provides the following constraints on the origin of the lavas: (i) the eruptions must have taken place very rapidly to prevent the xenolith from being absorbed by the host magma; (ii) the magmas probably originated from the mantle, because they contain xenoliths representative of all lithosphere layers; and (iii) brittle lithospheric fractures must have been present to allow magmas to ascend. Finally, the occurrence and geochemistry of olivine xenocrysts from site A lavas imply a derivation from the oceanic lithosphere at depths of ~14 km (*10*); the depth to the base of the crust in this location is 7 to 8 km (*11*).

The lava samples were alkalic with steep primitive mantle–normalized rare earth element (REE) patterns (*8*) (fig. S2 and table S2). This suggests that they formed from small degrees of melting in the presence of garnet, which retains the heavy REEs (>90 km deep) (*12*). In the standard model of oceanic plates, the 135 million-year-old Pacific Plate would be ~95 km thick (*4*); thus, the melting probably took place in the asthenospheric mantle beneath the Pacific Plate. The high vesicularity of the rocks was probably caused by the presence of $CO_2$, because it is not very soluble in alkalic magmas, in contrast to $H_2O$ (*8*, *13*).

The composition of the mantle source is important to determine in order to exclude a hot spot origin for the petit spot magmas. Mid-ocean ridge basalts (MORBs), presumed to be sourced from the upper, depleted asthenospheric mantle, have radiogenic noble gas isotopic compositions (for example, low $^3He/^4He$ ratios), whereas ocean island basalts (OIBs) have unradiogenic values. This is because nucleogenic, radiogenic, or fissiogenic isotopes of $^{21}Ne$, $^{40}Ar$, and $^{129,131-136}Xe$ are more abundant in the degassed MORB-source mantle than in the OIB-source mantle, which is generally less degassed and may even be undegassed (that is, primitive) (*14*, *15*). Our samples from site B are highly radiogenic in noble gas isotope compositions, suggesting a similar source to that of MORB (Fig. 2 and table S3) (*8*). The high $^{40}Ar/^{36}Ar$ ratios (>10,000) are higher than any reported for OIBs (*16*) and are convincing evidence for an upper mantle source.

A key question is whether the volcanoes represent a hot spot fed by a mantle plume. The presence of a tomographically imaged low-

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velocity region (a zone potentially representing a mantle plume) at a 410-km depth in this area was reported by Obayashi *et al*. (*17*); however, there is no evidence for a conduit or connection of any type between this low-velocity region and the shallower mantle. Our geochemical evidence strongly supports a depleted mantle (nonplume-like) source. Furthermore, the volume of magma produced at the petit spot volcanoes must have been several orders of magnitude less than those typical of hot spot volcanoes. We found 4.2 to 8.5 million-year-old volcanoes at site A, suggesting episodic eruption of magma over a distance of 400 km of plate motion. Accordingly, the petit spot volcanic province is characterized by several million years of small-volume magma production over a large area.

Temperatures of the asthenosphere beneath the Pacific Plate have been mapped precisely by means of surface wave tomography, showing that the thermal state of the asthenosphere is notably homogeneous throughout the Pacific Ocean (*18*). The temperature at a depth of 150 km is estimated to be between 1450° and 1480°C, which implies that the temperature just below the Pacific Plate is 50° to 150°C lower than the solidus of dry mantle materials (*19*). However, at this depth and temperature in the asthenosphere, a small percent melt should exist in the presence of small amounts (<1%) of $H_2O$ or $CO_2$ (*20*), which lowers the melting temperature. The highly vesicular petit spot lavas probably represent incipient partial melts that formed in the asthenosphere in the presence of volatiles, most plausibly $CO_2$. If the magmas were supplied from the normal asthenosphere, with emplacement channels controlled by tectonic fracturing of the overlying lithospheric plate, the low volumes of magma output over a large area could easily be explained.

An old and cold lithospheric plate behaves elastically and may be flexed because of loading by an ocean island or seamount or by subduction-related plate flexure (*21*). In the area between sites A and B, the bathymetric high (or outer rise) is aligned parallel to the Japan Trench. The Pacific Plate flexes convexly here, as it subducts beneath Japan and yields a positive gravity anomaly (*22*). This flexed region is elevated >800 m above the normal ocean floor (~6000 m below sea level at site B). Large curvatures imposed on the preflexed lithosphere might instigate brittle fracturing (that is, bending-induced faults) (*23*). As for volcanic cones at site A, the volcanic features, aligned in a west-northwest to east-southeast direction, are essentially perpendicular to hinge lines on the bending plate (Fig. 1, A to C). Accordingly, it appears that magmas are brought to the surface along fractures parallel to the direction of the maximum horizontal compression. However, the surface compression is actually caused by extensional stresses on the base of the downwarping Pacific Plate (Fig. 3C).

Post–erosional-stage lavas on some of the Hawaiian Islands, as well as submarine lavas on the flexural Hawaiian Arch (because of loading on the plate), have chemical compositions and tectonic emplacement mechanisms (*24*) that are similar to those of the petit spot lavas. It has also been proposed that alkalic lavas in the western Samoan Islands (located on the opposite side of the main Samoan hot spot shield volcanoes) may be derived from the asthenosphere during tectonic faulting (*25*). If true, a similar style of volcanism may be present

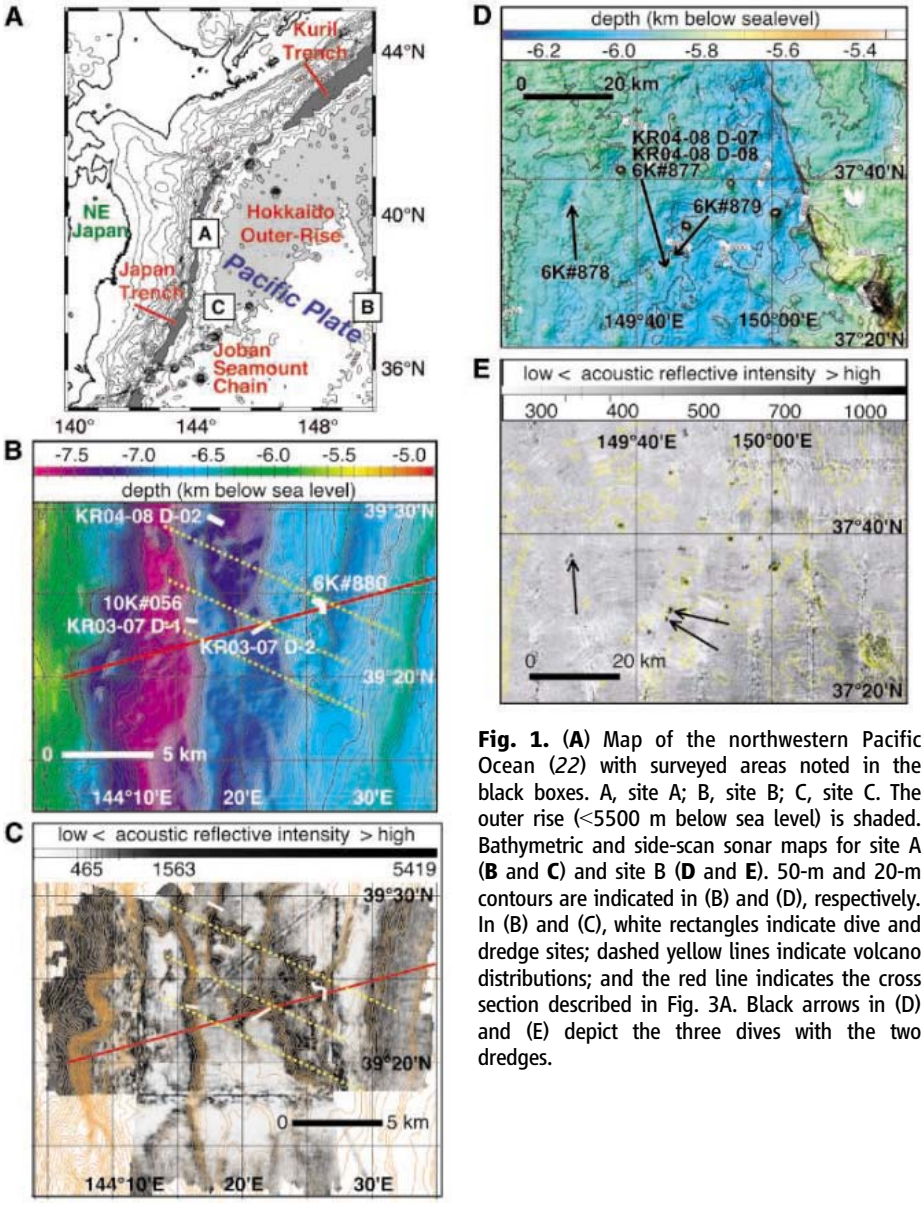


**Fig. 1.** (**A**) Map of the northwestern Pacific Ocean (*22*) with surveyed areas noted in the black boxes. A, site A; B, site B; C, site C. The outer rise (<5500 m below sea level) is shaded. Bathymetric and side-scan sonar maps for site A (**B** and **C**) and site B (**D** and **E**). 50-m and 20-m contours are indicated in (B) and (D), respectively. In (B) and (C), white rectangles indicate dive and dredge sites; dashed yellow lines indicate volcano distributions; and the red line indicates the cross section described in Fig. 3A. Black arrows in (D) and (E) depict the three dives with the two dredges.

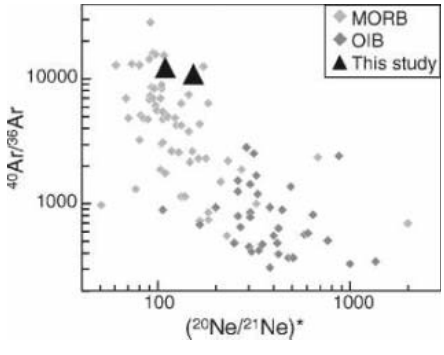


**Fig. 2.** Plot of Ne versus Ar isotopes. $(^{20}Ne/^{21}Ne)^*$ is equal to $[(^{20}Ne/^{22}Ne) - (^{20}Ne/^{22}Ne_{Air})]/[(^{21}Ne/^{22}Ne) - (^{21}Ne/^{22}Ne_{Air})]$, showing the slope of a mixing line on the conventional neon three-isotope plot ($^{20}Ne/^{22}Ne$ versus $^{21}Ne/^{22}Ne$) (*8*). Data with $^{20}Ne/^{22}Ne$ ratios of less than 10 were not plotted (*8*). Literature and data for MORB and OIB may be found in the following online databases: Petrological Database of the Ocean Floor (www.petdb.org/) and Geochemistry of Rocks of the Oceans and Continents (http://georoc.mpch-mainz.gwdg.de/georoc/).

on other oceanic plates where extensive flexure and fracturing are taking place. Investigating these types of areas may prove to be a fruitful endeavor and result in the detection of other small volcanic features. For example, we have also observed volcanic cones and/or highly reflective sonar images, similar to those observed at the petit spot, elsewhere on the Pacific Plate (site C in Fig. 1A); these small cones will be examined further in the near future.

Early in the theory of plate tectonics, the presence of partial melts in the asthenosphere was considered to correspond to the solidus of Earth's mantle (*1*, *26*). This hypothesis was supported by observations of a seismic low-velocity zone, a highly electric conductive layer, and an experimentally determined peridotite solidus in the presence of volatiles (*27*). More recently, the argument for the existence of partial melts in the asthenosphere has been weakened by studies, such as laboratory measurements of mantle material (*28*) and the predicted rheology of the mantle (*29*), that question its necessity. Therefore, this documentation of young alkali basalts on the old Pacific Plate provides new and strong support to the long-standing idea of the asthenosphere as a zone of partial melt.

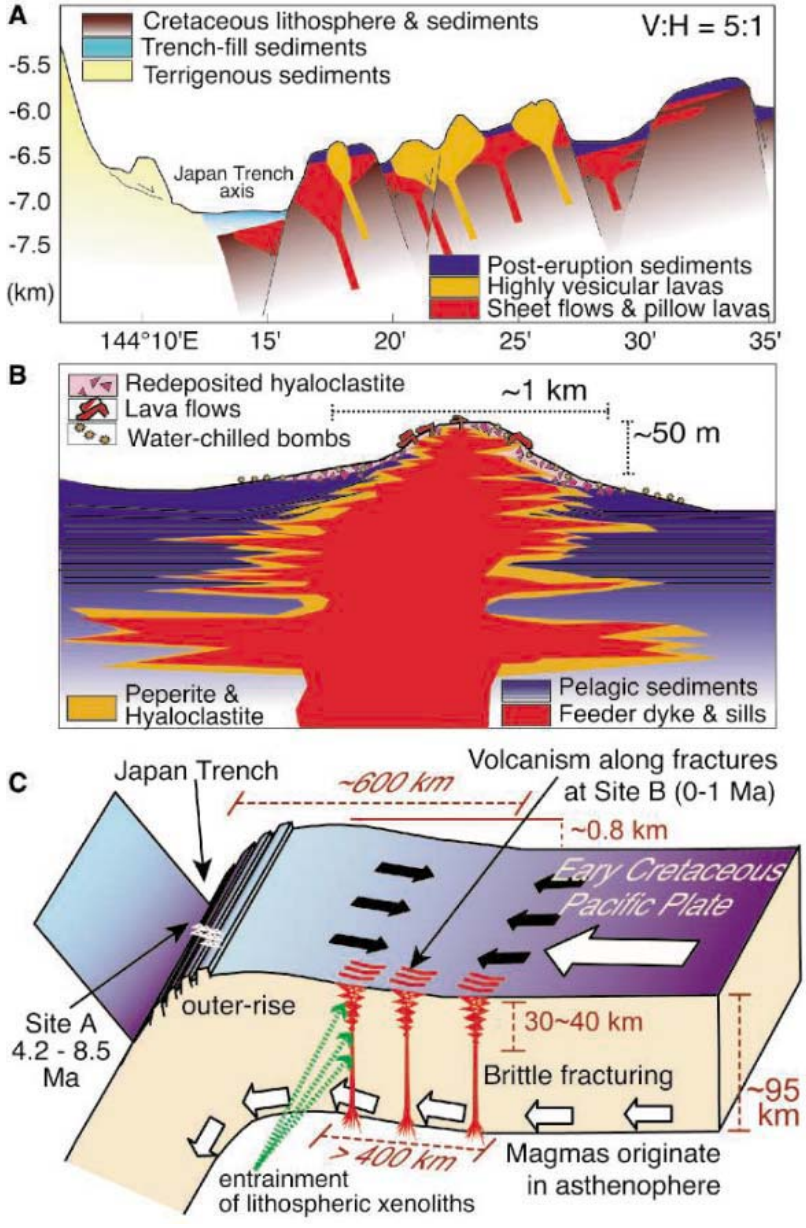


**Fig. 3.** Geologic interpretations of the lava fields. (**A**) Schematic cross section of site A (red line in Fig. 1, B and C). (**B**) A schematic cross section of a volcano at site B constructed from observations made during dives. (**C**) A conceptual model of petit spot volcanism. Magmas from the asthenosphere escape to the shallow depths because of the extensional environment of the lower lithosphere (*21*) and migrate up through the brittle compressed upper lithosphere by exploiting fractures created by flexure of the plate. Data for the plate and flexure framework are from previous studies (*6*, *11*, *21*, *22*).

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### Supporting Online Material

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# Cold-Seep Mollusks Are Older Than the General Marine Mollusk Fauna

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The origin and possible antiquity of faunas at deep-sea hydrothermal vents and seeps have been debated since their discovery. We used the fossil record of seep mollusks to show that the living seep genera have significantly longer geologic ranges than the marine mollusks in general, but have ranges similar to those of deep-sea taxa, suggesting that seep faunas may be shaped by the factors that drive the evolution of life in the deep sea in general. Our data indicate that deep-sea anoxic/dysoxic events did not affect seep faunas, casting doubt on the suggested anoxic nature and/or global extent of these events.

Deep-sea cold seeps are sites where low-temperature fluids rich in hydrocarbons and hydrogen sulfide leak onto the sea floor and support specialized animal communities dominated by taxa having symbiotic relationships with chemoautotrophic bacteria (*1*). The trophic structure of seep communities is similar to that of hydrothermal vent communities, and many of the dominant chemosymbiotic macrofauna are shared at species, generic, and family levels (*2*). Because of their reliance on an in situ chemosynthetic food source, these communities may have had an independent evolutionary history (*3*, *4*). It has been suggested that many vent and seep taxa underwent rapid bursts of adaptive radiation and extinction (*5*) or had Mesozoic or even Paleozoic origins and subsequent long-term in situ radiations (*3*, *4*, *6*, *7*). This latter antiquity hypothesis is based on the geologic age of sister groups to these taxa from nonchemosynthetic sites and the high degree of endemism at the family and even higher taxonomic level at vents and seeps. In contrast, molecular divergence date estimates calibrated against the fossil record of nonendemic taxa indicated late Mesozoic or Cenozoic origins for various other groups (*8*–*11*). This discrepancy has been attributed to oceanic anoxic/dysoxic events in the late Mesozoic and early Cenozoic that would have periodically extirpated most vent communities, with recolonization from putative shallow-water vent refuges (*10*–*12*), in which case seep communities should have been similarly affected. We tested the influence of these biologic and paleoenvironmental factors on the evolutionary history of the seep fauna using direct fossil evidence.

One hundred and two bivalve, gastropod, and polyplacophoran genera are recorded from 125 fossiliferous seep sites ranging from Jurassic to Recent (*13*). No mollusk taxa have been identified to the genus level at seeps older than the late Jurassic. Our definition of seep genera includes endemics and symbiotic and nonsymbiotic genera, but not transient taxa, which are mostly removed by excluding 34 fossil single-occurrence genera. Also excluded from our analyses are 32 modern seep genera without fossil occurrences at seeps. The resulting range chart includes 36 genera, 29 of which occur in extant seeps (Fig. 1). Of the extant genera, 13 are vent, seep, and whale-fall (living on a whale carcass that has fallen to the ocean floor) endemics. Common and widespread taxa are better represented in the fossil record of seeps than are rare taxa. Of the 61 extant seep genera, 62% (38 genera) occur only at a single locality or in a narrow geographic area. Among the 32 extant seep genera without a fossil record, 78% (25 genera) occur only at a single locality and are usually rare there. Contrary to previous assumptions (*14*), no size-related bias was detected in the fossil record of seep mollusks. Among the living seep genera, 20% have shells smaller than 5 mm in height, length, or width among fossil genera, the value is 16% for the same size class.

To assess the evolutionary age of the living seep genera, the frequency distribution of the first occurrence of the 29 extant seep genera with a fossil record, sorted into eight geologic bins from the Late Jurassic to the Plio-Pleistocene, was compared to the frequency distribution of the first occurrence of the 2366 living bivalve, gastropod, and polyplacophoran genera (*15*), sorted into the same time intervals. The fact that these time intervals have different durations does not affect the analysis, as we are comparing like with like. The two frequency distributions (Fig. 2A) have significantly different medians (Wilcox test, $W = 49596$, $P < 0.00005$). A randomization test using the same test statistic and 100,000 replicates showed that this result is significant at the 0.001% level (*13*). The median of the seep genera is in the Eocene and that of the marine genera is in the Oligocene. Thus, seep genera are on average one geologic series older than the general marine mollusk fauna, which does not make them a relic fauna. New taxa appeared at seeps in each time interval since the late Jurassic, except for the Paleocene. Dating uncertainties and the uneven temporal distribution of seeps do not allow continuous or punctuated patterns to be distinguished. There are no indications of long-term in situ radiations. Of 36 genera in the analysis, 15 are endemic and belong to 11 families, 6 of which are themselves endemic (*13*).

Sampling bias could be a potential source of error in our analysis because of the positive correlation between available outcrop area and taxon diversity (*16*, *17*). This bias may be operating in the Mesozoic and Paleogene, where there is a rough correlation between the frequency of new taxa appearing and the abundance of seep-bearing formations (Fig. 2B). However, it does not hold true in the Neogene, a time period that contains more than half (54%) of all seep-bearing formations but only 10% of seep genera first occurrences (*13*), suggesting that the small number of first occurrences in the late Cenozoic is a true biological signal. An alternative hypothesis not invoking geologically older seep mollusks is that the frequency distribution of first occurrences among the general marine fauna is skewed toward a younger age by a few genera-rich families that have radiated recently, whereas the seep genera belong to the remaining older general marine fauna. To assess this hypothesis, we compared the frequency distribution of first occurrences of the general marine fauna to that of the five most genera-rich mollusk families and superfamilies (gastropods Buccinidae, Muricidae, and Turridae; bivalves Tellinoidea and Veneroidea). These taxa constitute 15% of the total number of marine mollusks (*13*), and the frequency distributions of their first occurrences are almost identical to that of the general marine fauna. We therefore reject the "recent taxon skew" hypothesis.

Organisms living at seeps may have been buffered from mass extinctions that affected marine faunas in general because, being fueled by in situ chemosynthetic production, photic zone productivity and food-chain disruption should have had little effect (*3*). Our data indicate a proportionally high number of first occurrences in the Cretaceous as compared to the general marine mollusks (Fig. 2A), and these taxa passed unaffected through the end-Cretaceous extinction event (Fig. 1). However, given the paucity of Maastrichtian and Paleocene seep sites, a thorough evaluation of the effects of the end-Cretaceous extinction on the seep fauna is not yet possible. A similar lack of effect is seen at the four largest anoxic/dysoxic events since the late Mesozoic (Fig. 1). Therefore, we doubt that these events had a significant influence on the evolution of deep-sea vent and seep faunas.

Based on shared species, it was hypothesized that whale falls could have acted as dispersal stepping stones for vent and seep taxa, and that the appearance of large ocean-going whales facilitated radiation into these environments (*18*, *19*). A high number of first occurrences of seep taxa is coincident with the appearance of basilosaurids in the middle to late Eocene (Fig. 2A), the first group of whales to have a global distribution (*18*). This appears to support the stepping-stone hypothesis, but the lack of deep-water records of basilosaurids raises ques-

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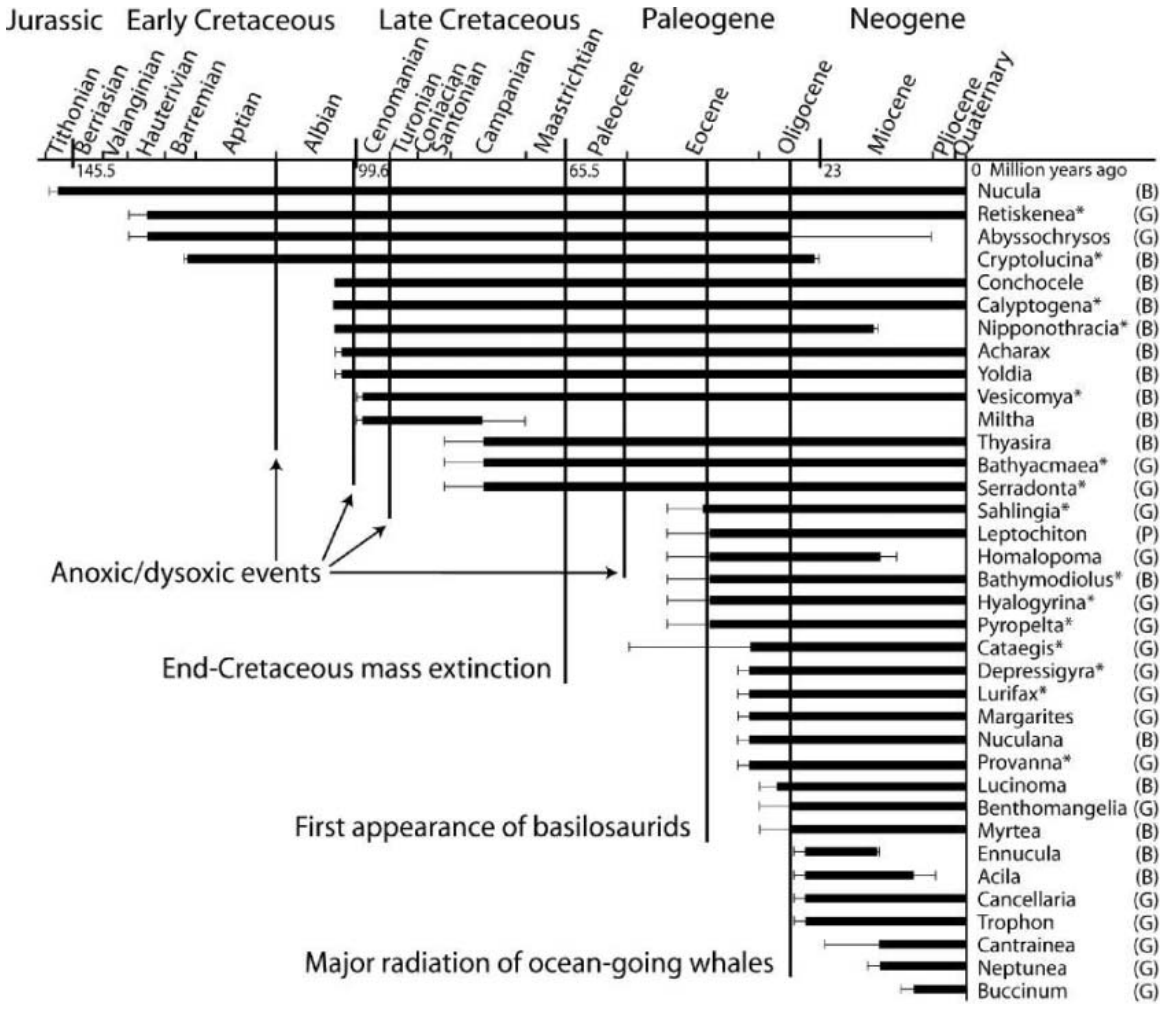


**Fig. 1.** Geologic ranges of Recent and fossil seep mollusks. Range bars start at the arithmetic middle of the published time interval of the respective localities; error bars indicate the published maximum range. (B), bivalve; (G), gastropod; (P), polyplacophoran. Asterisk indicates a genus endemic to vents, seeps, and whale falls.

tions about the ocean-crossing abilities of this group (*20*). The major radiation of unequivocal ocean-going whales in the mid-Oligocene (*21*) is synchronous with the first fossil whale-fall communities (*22*), but because 76% of all seep mollusks originated before that time (Figs. 1 and 2A), a movement of taxa from seeps to whales seems the more likely scenario.

The deep sea generally holds a higher percentage of geologically old taxa than do shallow marine environments, resulting from onshore-offshore patterns in the evolution of marine organisms (*23*, *24*). Therefore, the older-than-average age of seep mollusks may be an extension of that seen in the deep sea (*3*). A comparison of first occurrence data on seep genera to those on mollusk genera that largely live in the deep sea (*13*) shows no significant difference in median values (Wilcox test, $W = 1011$, $P = 0.12$). This may indicate that the factors driving the evolution and survival of deep-sea taxa affect the seep fauna as well, regardless of their independent food source. Modeling of taxon-age relationships has shown that low origination rates can, in some

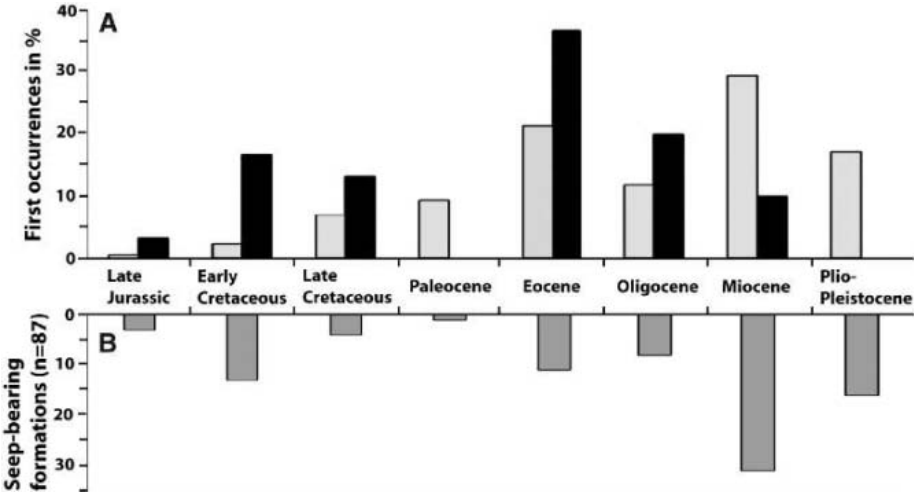


**Fig. 2.** Frequency distributions of the first occurrence of seep mollusks, of marine mollusks in general, and the number of seep-bearing formations from the late Jurassic to Recent. (**A**) First occurrence data of seep genera (black bars, $n = 29$) against those of marine mollusks (light gray bars, $n = 2366$), shown in percent of the total number of genera in each data set. The time of first occurrence corresponds with the onset of the solid range bars in Fig. 1. (**B**) Number of seep-bearing formations for the same time intervals.

cases, result in longer taxon ranges than high origination rates (*25*). The paucity of seep taxon origination in the Neogene could indeed reflect low origination rates in the seep environment and thus account (to some extent) for the older-than-average age of seep mollusks. However, deep-sea mollusks show high origination rates in the Neogene, despite their older-than-average age, and considering the lack of effect of the end-Cretaceous extinction event and the anoxic/dysoxic events on both deep-sea and seep taxa, we conclude that paleoenvironmental factors have mainly shaped the age distribution of the modern seep mollusks.

Vents and seeps have been considered extinction-resistant habitats (*3*, *4*). Extinction and survival patterns across the Paleocene/Eocene thermal maximum show a paradox. Planktonic foraminifera and calcareous nannofossils remained abundant, but a significant number of benthic foraminifera became extinct, suggesting a cause that preferentially affected the deep-sea biota (*26*). However, there was no extinction among the seep fauna, deep-sea echinoids (*27*), and higher-level taxa among vent organisms (*28*). Because seep taxa are a mixture of endemics and deep-sea generalists, more work is needed to disentangle whether their apparent resistance to extinction is a seep-related phenomenon or a trait shared with deep sea organisms in general.

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### Supporting Online Material

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References

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# Temporal and Spatial Enumeration Processes in the Primate Parietal Cortex

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**Humans and animals can nonverbally enumerate visual items across time in a sequence or rapidly estimate the set size of spatial dot patterns at a single glance. We found that temporal and spatial enumeration processes engaged different populations of neurons in the intraparietal sulcus of behaving monkeys. Once the enumeration process was completed, however, another neuronal population represented the cardinality of a set irrespective of whether it had been cued in a spatial layout or across time. These data suggest distinct neural processing stages for different numerical formats, but also a final convergence of the segregated information to form most abstract quantity representations.**

Humans and animals share an evolutionarily ancient quantification system that allows them to approximately estimate the size of a set (its numerosity) without verbal symbols (*1*–*4*). How numerical information can be extracted depends on the presentation format: whether the elements of a set are perceived simultaneously or sequentially. When presented simultaneously as in multiple-item patterns, numerosity can be estimated from a spatial arrangement at a single glance. Here, parallel processing mechanisms are engaged, as indicated by constant reaction times (*5*–*7*), equal numbers of scanning eye movements (*7*), and comparable neural response latencies (*8*) across absolute set size. In contrast, when the elements of a set are presented one by one, they need to be enumerated successively across time (*9*–*11*). This latter process constitutes a nonverbal precursor of real counting, which is a sequential enumeration process via number symbols.

Both human (*12*–*21*) and monkey (*22*, *23*) studies relate the processing of numerical quantity information to the posterior parietal cortex, including the intraparietal sulcus (IPS). However, none of these studies addresses the actual "counting" aspect, namely the abstract accumulation of sensory events one by one. Moreover, it remains unknown whether and how numerical information extracted from temporally and spatially arranged presentations is combined at the neuronal level.

To investigate the role of individual IPS neurons in representing simultaneously and sequentially presented visual quantity, we trained monkeys to discriminate small numerosities. The monkeys had to judge whether two successive task periods (first sample, then test) separated by a 1-s delay contained the same number of items (one to four) (Fig. 1, A and B). If so, the animals had to release a lever. In the sample period, the numerosity was presented randomly: either by multiple-dot patterns showing the number of items simultaneously (the simultaneous protocol, Fig. 1B) or by single dots appearing one by one to indicate the number of items in sequence (the sequential protocol, Fig. 1A) (*24*). We ensured that non-numerical parameters (visuospatial cues in multiple-dot patterns and temporal cues in the sequential presentation protocol) could not be used by the monkeys to solve the task. Controls in the simultaneous protocol included displays in which the circumference (and with it total area), density, and configuration (shapelike or linear) were controlled across different quantities. Controls in the sequential protocol eliminated temporal factors that may covary with increasing numbers of sequential items, such as the total duration of the sample period, the duration of individual items and pauses in between, the total visual energy (or total area across time, respectively), and the regularity (rhythm) of the item sequence (see Table 1 and fig. S1 for details). In the test period, numerosity was always cued by multiple-item displays.

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The monkeys were first trained on the simultaneous task alone and subsequently on the sequential numerosity discrimination. Initially, they only learned to discriminate sequential numerosity 2 from 4 (and vice versa). To investigate whether the monkeys would understand the concept of sequential numerosity without further training, we introduced transfer tests requiring the monkey to discriminate sequential numerosity 3 from 2 and 4 (see SOM for details). Because the animals were randomly rewarded for their responses in transfer tests, any learning of the "correct" response was impossible. Both monkeys immediately discriminated sequential numerosity 3, with a precision comparable to the baseline discrimination of 2 versus 4 (as indicated by the fact that the slopes for the match and nonmatch discrimination in the transfer and baseline tests are almost parallel) (Fig. 1, C and D). The numerical distance between match and nonmatch in the transfer tests is only one, which is more difficult to discriminate than the baseline discrimination with a numerical distance of two (*25*).

After these transfer tests, monkeys had to perform both the simultaneous and the sequential task within a single session. The average performance of the two monkeys on both the sequential and simultaneous protocols was between 71 and 99% correct (Fig. 1E) and significantly better than chance for all tested quantities (binomial test, $P < 0.001$; see fig. S2 for detailed performance curves). Discrimination of the quantity of sequentially presented items was more difficult for both monkeys (binomial test, $P < 0.01$). The monkeys readily generalized to the control stimulus sets; performance was very similar across them in the sequential and simultaneous protocols (Fig. 1E and fig. S2).

We recorded from 228 randomly selected neurons in the depth of the IPS (Fig. 2, A and B) of two monkeys performing the numerosity discrimination task. Random presentation of either the sequential or simultaneous protocol from trial to trial allowed us to investigate an individual neuron's responses to both presentation types in an unbiased way. A proportion of the tested neurons [27 out of 228 (27/228) or 12%, two-way analysis of variance (ANOVA), with factors (stimulus protocol) × (sample numerosity), $P < 0.01$] showed activity that varied significantly with the number of items during sample presentation in the simultaneous protocol, irrespective of the displays' visuospatial properties (*23*). Even more neurons (58/228 or 25%, two-factor ANOVA, $P < 0.01$) showed maximum discharge in response to a certain number of dots in the sequential protocol. To further test whether the neurons' discharges to the preferred sequential item were unaffected by temporal parameters, we computed a multiple regression analysis with the spike rate to the preferred sequential item as a dependent variable and the duration of the preferred item in a sequence, the duration of the pause preceding the preferred item, and the duration of the previous-to-preferred item as independent variables (*24*). Only 8 of the 58 sequential numerosity-selective neurons exhibited a significant correlation ($P < 0.01$) with temporal parameters and were thus excluded from the pool of numerosity-selective neurons (fig.

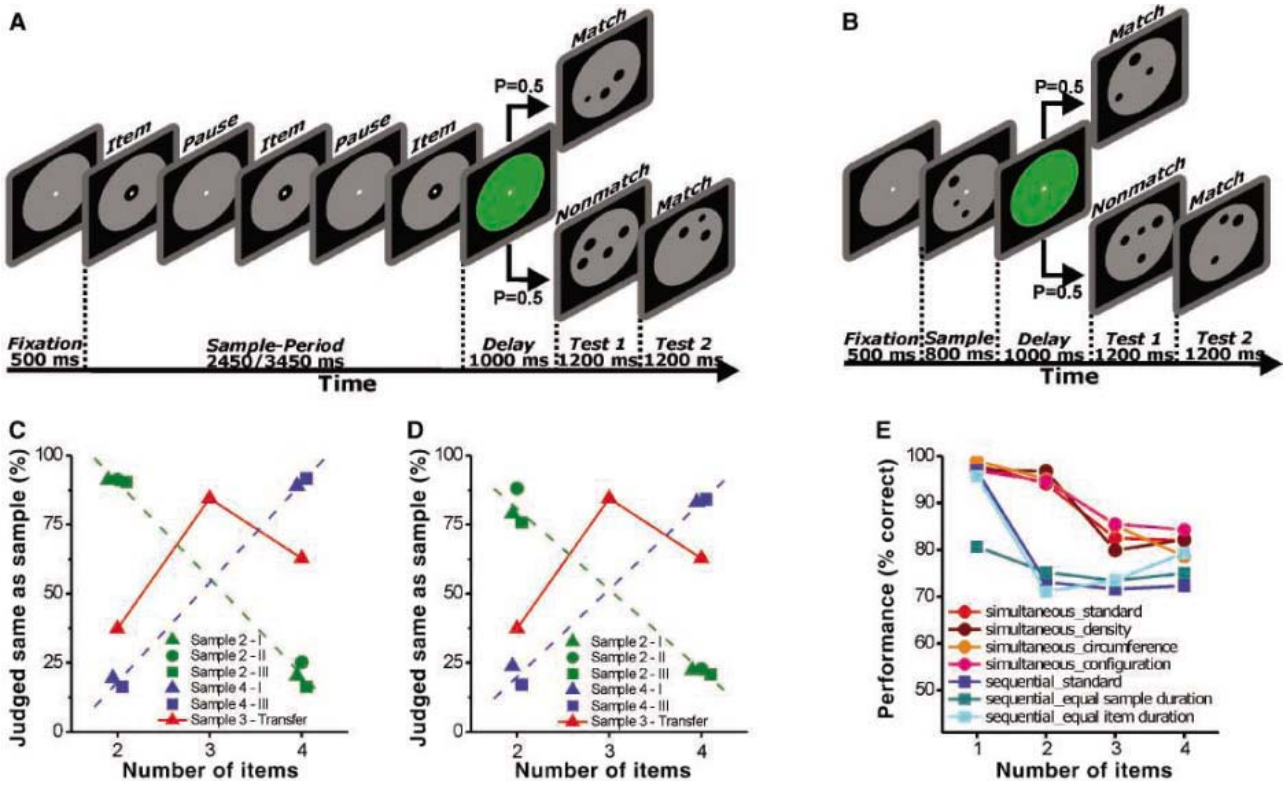



**Fig. 1.** Task protocols and behavioral performance. (**A**) Sequential delayed match-to-quantity task (for example, numerosity 3). A trial started when the monkey grasped a lever. The monkey had to release the lever if the sample period and test display contained the same number of items but continue holding it if they did not (probability of match/nonmatch condition = 0.5). The sample numerosity was cued by sequentially presented items temporally separated by pauses containing no items. The temporal succession and duration of individual items were varied within and across quantities (Table 1). The numerosity in the test period was always cued by multiple-item displays. (**B**) Simultaneous delayed match-to-quantity task. Task conditions were identical to those in the sequential protocol, but numerosity was cued by a single multiple-dot display in the sample period. The physical appearance of the displays varied widely for the same quantities (Table 1). For both the simultaneous and sequential task protocols, the nonmatch stimuli for sample numerosity 1 was 2; for sample 2 the nonmatch numerosities were 1 and 3 (probability = 0.25); for sample numerosity 3 and 4, nonmatches were one and two numbers up and down. (**C** and **D**) Behavioral performance of both monkeys during transfer tests in the sequential task. The monkeys performed a baseline discrimination of 2 versus 4 [green data for different stimulus protocols (I to III) are displayed as listed in Table 1] and 4 versus 2 (blue). Both monkeys [(C) and (D)] spontaneously discriminated sequential numerosity 3 from 2 and 4 in transfer tests without reinforcement (red). (**E**) Average performance of both monkeys in the simultaneous and sequential tasks (under standard and control conditions) during the recording sessions. Chance performance = 50% (also see fig. S2 for details).

S4). Based on the combined results from the two-factor ANOVA and the multiple regression analysis, 50 neurons (from a total of 228 cells; that is, 22%) were found to be selective to sequential quantity only. Two example neurons tuned to sequential numerosity are shown in Fig. 2. The tuning function of the neuron in Fig. 2, C to E, showed peak activity for the sequential quantity 2 and a systematic dropoff of activity as the number of items in the sample period varied from the preferred value (Fig. 2D); this was true even in trials with three or four consecutive dots and varied sequence timing (Fig. 2E). A neuron with preferred numerosity 4 is plotted in Fig. 2, F to H.

Similar response profiles were observed for all neurons tuned to numerosities 1, 2, 3, or 4 in the sequential (see population tuning curves in Fig. 3A) or simultaneous (Fig. 3B) protocol. Each cell showed peak activity for one of the visual quantities and a systematic dropoff of activity as the number of sample items varied from the preferred value (Fig. 3C). Numerosity 1 was the most frequent preferred numerosity in both protocols (Fig. 3D).

Many of the tested neurons (43/228 or 19%) also showed activity that varied significantly during the pauses between item presentation in the sequential protocol (Fig. 1A), irrespective of the temporal arrangement [again tested with a combination of a two-factor ANOVA and a multiple linear regression analysis; see supporting online material (SOM) and fig. S5 for details]. More than half of the neurons that were significantly tuned during the pauses were also tuned during sequential item presentation (25/228 or 11%); thus, the firing rates during item presentations as well as during the pauses between them varied with the position in the sequence. The neuron in Fig. 2F, for example, exhibits a significant increase in discharge during successive pauses, with the highest discharge during the third pause in the sequence. At the same time, this neuron has the preferred item position "four" (Fig. 2, G and H). We found a significant correlation between the neurons' preferred serial position during the pauses and the preferred number of sequential items ($r = 0.70$, $P < 0.001$, $n = 25$ neurons), indicating that the activation of neurons between items could provide a neuronal storage mechanism to keep track of the actual items to enumerate. Such an accumulation of activation was successfully implemented in neural networks simulating numerosity detection (*26*).

A comparison of neurons tuned to numerosity during the sample period in the sequential and simultaneous protocols showed a clear dissociation of neuronal populations. Only 10 neurons (4% of the total sample) were significantly tuned to numerosity in both protocols. Of those 10 cells, only 2 (1% of the total sample) were tuned to the identical numerosity in both the sequential and simultaneous protocols (see neuron in Fig. 2F and fig. S3), which corresponds to chance occurrence. This finding suggests that different populations of neurons are engaged in extracting numerosity in a parallel or serial fashion, respectively (Fig. 3E).

Only after the enumeration process in the sample period was completed did the animals have full information about the cardinality of a set. They had to maintain the quantity in mind throughout a delay period and prepare to find the matching quantity in a test display. Many neurons (43/228 or 19%) were significantly tuned to numerosity only in the memory period, irrespective of the presentation protocol (only significant numerosity effect, two-factor ANOVA with numerosity and presentation protocols as factors, $P < 0.01$). For example, the neuron displayed in Fig. 4 showed remarkably similar delay activity in the sequential (Fig. 4A) and simultaneous (Fig. 4B) presentation protocol, with 3 as the preferred numerosity (Fig. 4C). The average response profiles of all numerosity-selective neurons during the delay period are shown in Fig. 4D. Few cells (9/228 or 4%) were tuned to numerical quantity but also differentiated between the simultaneous and sequential presentations (numerosity and presentation protocol effect or interaction, two-factor ANOVA, $P < 0.01$). Some neurons tuned to the set size in the delay period represented quantity in the sample period (simultaneous protocol: 7/43 or 16%; sequential protocol: 9/43 or 21%).

An examination of error trials suggested that the delay activity of IPS neurons was directly related to the monkeys' performance. When monkeys made judgment errors, neural delay activity for the preferred numerosity was significantly reduced to 83.6% of that observed on correct trials (i.e., 100%) ($P = 0.01$, Wilcoxon signed ranks test, two-tailed).

These results argue for segregated processing of simultaneous and sequential numerical quantity. Different populations of neurons were involved in extracting numerosity across spatial or temporal arrangements during an ongoing quantification process. In contrast, the final and common result of the quantification process was coded by a third population of neurons, irrespective of whether numerosity was cued simultaneously or in sequence. Thus, the intermediate numerosity of an ongoing quantification process and the storage of the final cardinality are accomplished by different neuronal populations.

In contrast to a direct, perceptual-like assessment of numerical information in multiple-item displays, sequential enumeration requires a more complex coding of numerical information. Our data point toward the pauses between individual successive items as a potential key mechanism for the coding of sequential numerosity. For many neurons, activation changes during the pauses were sometimes more prominent than during the presentation of successive items (Fig. 2F). In these neurons, activation to the item presentation seemed to ride on ever-increasing discharges during the pauses, which is consistent with the idea of an accumulator mechanism (*9*, *26*).

The enumeration of sequentially presented items requires an organism to keep track of the serial position of the previously presented item. Similar to our findings in nonhuman primates, neuropsychological (*27*) and electrophysiological

**Table 1.** Stimulus presentation protocols and variation of non-numerical parameters with quantity (w.q.). The timing of the sample period in the sequential protocol was as follows: Standard protocol: sample period duration for all numerosities, 2450 ms; single item/pause duration for numerosity 1, 409 to 1328 ms; numerosity 2, 372 to 1359 ms; numerosity 3, 229 to 1066 ms; numerosity 4, 212 to 607 ms. Equal sample duration protocol: sample period duration for all numerosities, 2450 ms; single item/pause duration for numerosity 1, 2450 ms; numerosity 2, 816 ms; numerosity 3, 490 ms; numerosity 4, 350 ms. Equal item/pause duration protocol: single item/pause duration for all numerosities, 350 ms; sample period duration for numerosity 1, 350 ms; numerosity 2, 1050 ms; numerosity 3, 1750 ms; numerosity 4, 2450 ms. For trials with a sample period duration of 3450 ms, values were correspondingly adjusted.

**Simultaneous protocol**

| Stimulus type | Spatial layout | Surface area | Circumference | Density |
|---|---|---|---|---|
| Standard | Randomized† | Increasing w.q. | Increasing w.q. | Increasing w.q. |
| Circumference | Randomized† | Decreasing w.q. | Equal | Increasing w.q. |
| Density* | Randomized† | Increasing w.q. | Increasing w.q. | Equal |
| Configuration | Linear | Increasing w.q. | Increasing w.q. | Increasing w.q. |

**Sequential protocol**

| Stimulus type | Sample period duration | Individual item or pause duration | Regularity (rhythm) | Intensity over time |
|---|---|---|---|---|
| Standard | Constant | Decreasing w.q. | Irregular | Variable |
| Equal sample duration | Constant | Decreasing w.q. | Regular | Decreasing w.q. |
| Equal item/pause duration | Increasing w.q. | Constant | Regular | Increasing w.q. |

*Density was determined by calculating the average distance between the dots. †High probability that three dots were arranged as a triangle, four dots as a quadrangle, and five dots as pentagon.

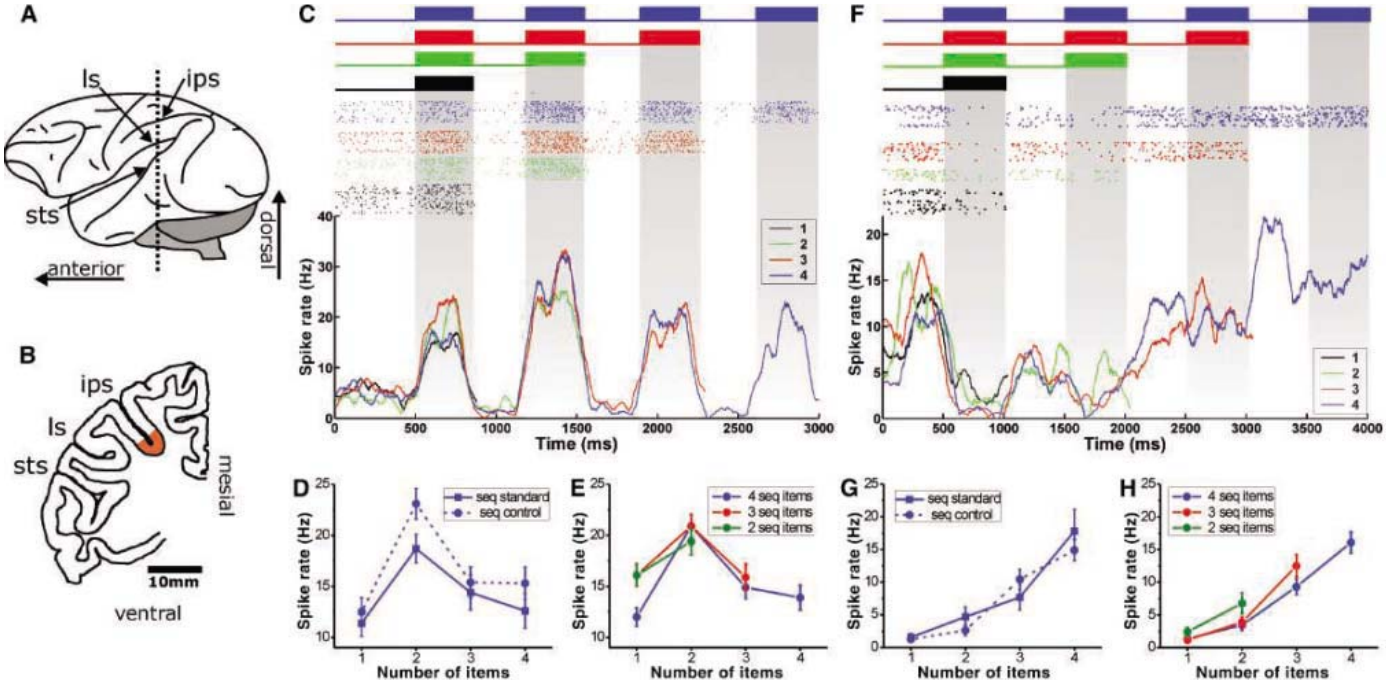
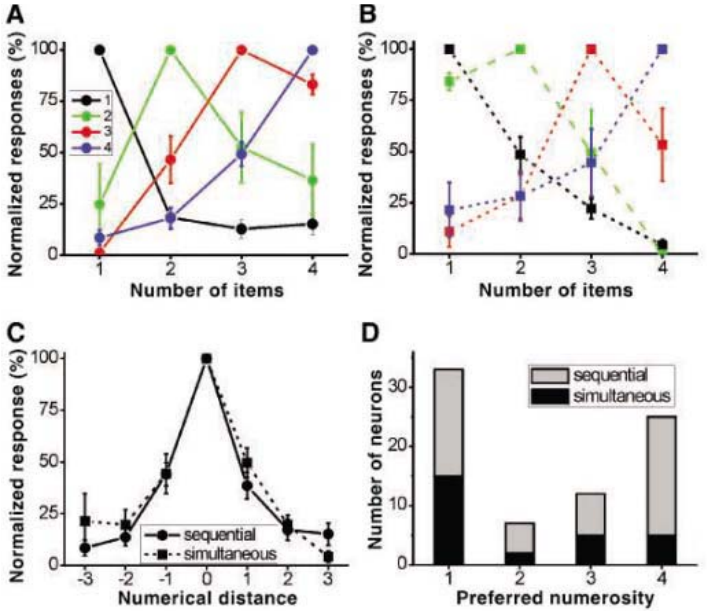



**Fig. 2.** Recording sites and neural responses during sample presentation. (**A**) Lateral view of the left hemisphere of a monkey brain indicating the topographical relationships of cortical landmarks. ips, intraparietal sulcus; ls, lateral sulcus; sts, superior temporal sulcus. (**B**) Coronal section at the level of the dotted line in (A) reconstructed from a structural magnetic resonance imaging scan (Horsley-Clark coordinates 0 mm anterior/posterior). The colored area in the depth of the IPS marks the recording area. (**C** to **E**) Responses of an example neuron selective to the sequential quantity 2 (only the "equal item/pause duration" protocol is shown for clarity). The top panel illustrates the temporal succession of individual items (square pulses represent single items). The corresponding latency-corrected discharges for many repetitions of the protocol are plotted as dot-raster histograms (middle panels; each dot represents an action potential) and averaged spike-density functions (bottom panels; activity averaged and smoothed). The first 500 ms represent the fixation period. Colors correspond for the stimulation illustration and the plotting of the neural data. Gray shaded areas represent item presentation. (D) Rate functions indicate the mean activity of the neuron in (C) to the standard and equal item/pause duration protocols [error bars in (D), (E), (G), and (H) represent the standard error of the mean] for four sequential dots. In both protocols, the neuron was tuned to numerosity 2. (Responses to the first item in a sequence of one, two, three, or four items were not statistically different.) (E) The same neuron shown in (C) was significantly tuned to numerosity 2 irrespective of whether the sample period showed two, three, or four sequential items (standard and control protocols pooled). (**F** to **H**) Neuron tuned to sequential numerosity 4. (F) Neuronal responses for the control protocol [layout as in (C)]. (G) Rate functions show monotonic increase of discharges up to numerosity 4 for both protocols. (H) Comparable discharges of this neuron to the sequential items, irrespective of whether the items were presented in sequences of two, three, or four items.



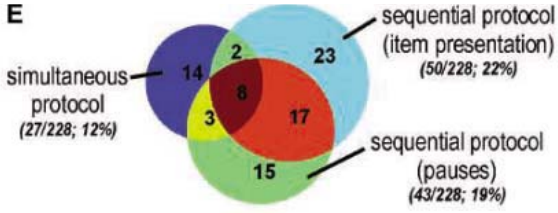




**Fig. 3.** Normalized responses averaged for neurons preferring the same sample quantity for the sequential (**A**) and simultaneous (**B**) protocols, respectively. (**C**) All neurons showed a progressive dropoff of the response with increasing numerical distances from the preferred numerosity, resulting in averaged tuning functions that were comparable for neurons tuned to sequential or simultaneous presentation. In (A) to (C), error bars represent the standard error of the mean. (**D**) Frequency distributions of the preferred numerosities for the sequential and simultaneous protocols. (**E**) Venn diagram illustrating the distributions of numerosity-selective neurons in the sample period. Some of the neurons exhibiting numerosity selectivity in the simultaneous protocol were also tuned in the sequential protocol during item presentation and/or during the pauses (indicated by graphical overlap), albeit mostly with different preferred numerosity. Numbers indicate the number of neurons per set.

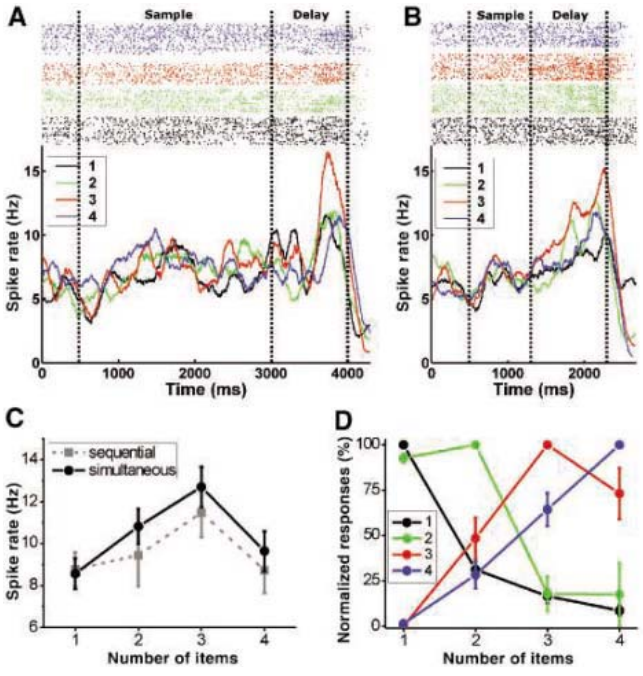



**Fig. 4.** Neural responses during the delay period. (**A** to **C**) A single neuron showing remarkably similar delay activity in the sequential (A) versus simultaneous (B) presentation protocol, with 3 as the preferred numerosity. Top panels in (A) and (B) show color-coded dot-raster histograms; bottom panels are the corresponding spike-density histograms. The first 500 ms represent the fixation period. This neuron was not numerosity-selective in the sample period. (C) Tuning function of the displayed neuron based on averaged discharge rates calculated over the delay period. (**D**) Normalized responses averaged for all neurons preferring the same quantity in the delay period, irrespective of the presentation protocol (sequential and simultaneous). In (C) and (D), error bars represent the standard error of the mean.

(*28*) studies in humans suggest dissociated processes involved in judging cardinality (numerical quantity) as opposed to ordinality (serial position), but sometimes with a common activation in the parietal and prefrontal cortices (*14*, *28*). Neurons in the lateral prefrontal cortex of the monkey are also selectively tuned to numerical rank (*29*) and numerical quantity (*8*), but typically later than IPS neurons (*23*). This suggests that neurons in the posterior parietal and prefrontal cortices are linked to form a single functional network for the representation of numerical information across space and time.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**Supporting Online Material**

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# Isolated Chloroplast Division Machinery Can Actively Constrict After Stretching


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Chloroplast division involves plastid-dividing, dynamin, and FtsZ (PDF) rings. We isolated intact supertwisted (or spiral) and circular PDF machineries from chloroplasts of the red alga *Cyanidioschyzon merolae*. After individual intact PDF machineries were stretched to four times their original lengths with optical tweezers, they spontaneously returned to their original sizes. Dynamin-released PDF machineries did not retain the spiral structure and could not be stretched. Thus, dynamin may generate the motive force for contraction by filament sliding in dividing chloroplasts, in addition to pinching-off the membranes.


All life depends on photosynthesis by chloroplasts in plants for food and oxygen. Chloroplasts arose from an endosymbiotic cyanobacterial ancestor and have their own genomes that are maintained by division (*1*). Electron-dense rings, designated the outer and inner plastid-dividing (PD) rings, are found on the cytosolic and stromal faces of the membranes at the equator of dividing chloroplasts and are thought to be ubiquitous throughout the plant kingdom (*2*). The outer PD ring is composed of a bundle of fine filaments 5 to 7 nm in diameter and is most likely to be associated with the generation of the constriction force through sliding of the filaments (*2*, *3*). In addition, two types of guanosine triphosphatases (GTPases), FtsZ and dynamin, are thought to participate in chloroplast division. FtsZ is a nuclear-encoded homolog of a key bacterial division protein (*4*) and forms a ring on the stromal side at the equator (*5*), whereas dynamin is a eukaryote-specific membrane fission protein (*6*–*8*) and forms a ring at the cytosolic side alongside the PD ring (*9*, *10*). Chloroplast division is thought to be controlled by a PDF


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machinery that includes a chimera of the rings from bacteria and eukaryotes, but the role of each ring in the PDF machinery is unclear.

The cells of the unicellular red alga *Cyanidioschyzon merolae* contain a single chloroplast per cell with an enormous PDF machinery. To isolate PDF machineries from *C. merolae*, dividing chloroplasts were obtained from synchronized cells (*9*) and treated with Nonidet P-40 (NP-40) and *n*-octyl-β-D-glucopyranoside (OG). The purity of the isolated PDF machineries was examined by fluorescence and electron microscopy, SDS–polyacrylamide gel electrophoresis (PAGE), matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS), and immunoblotting analysis. Immunofluorescence microscopy with antibodies to CmFtsZ2 (FtsZ) and to CmDnm2/DRP5 (dynamin) revealed that FtsZ and dynamin rings were localized to the constriction sites of chloroplasts in whole cells, as well as in isolated chloroplasts, from the early to late phases of chloroplast division (Fig. 1A). The FtsZ, dynamin, and outer PD rings behaved in a very similar way, even when the bulk of the chloroplast contents (thylakoid membrane with phycobilisomes and inner membrane) was dissolved by NP-40 treatment (Fig. 1, B and C). Subsequently, detergent treatment with OG for 5 min led to dissolution of the outer membrane. As a result, the membrane-free PDF machineries formed supertwisted rings, circular rings, and spirals (Fig. 1D). In addition, when isolated PDF-machinery fractions with or without the outer membrane were analyzed by SDS-PAGE and MALDI-TOF-MS, dynamin was not detected in the fraction with the outer membrane (NP-40 treatment), but was present in the fraction without the outer membrane (NP-40 and OG treatments) (Fig. 1E). Immunoblotting analysis indicated that dynamin and FtsZ were concentrated in the isolated PDF-machinery fraction (Fig. 1F). Thus, the isolated PDF machinery was intact, and it was composed of an FtsZ ring (inside) and PD and dynamin rings (outside) through the membranes.

Because the circumferences or lengths of isolated supertwisted (clockwise or anticlockwise spiral) PDF machineries (2.6 to 5.7 μm) were longer than those of circular PDF machineries (0.8 to 2.6 μm), the supertwisted and circular PDF machineries were considered to be derived from dividing chloroplasts at the early and late phases, respectively (Fig. 1D; figs. S1 and S2). Spirals arose from the supertwisted PDF machineries depending on the membrane solubilization by OG (Fig. 1D; figs. S1 to S3). The existence of supertwists and spirals suggested that the PDF machineries generate the driving force for contraction at the early phase of chloroplast division, thereby posing the question of which rings and/or factors generate the force in the PDF machineries (*6*, *11*). The presence of conformational changes could not be confirmed by the guanosine triphosphatase (GTPase) assay (fig. S4). However, when the NP-40–insoluble fractions were treated with OG for 5 min, FtsZ- or dynamin-released PDF machineries, in addition to intact PDF machineries, were obtained. The FtsZ-released and intact PDF machineries showed spiral structures, whereas the dynamin-released PDF machineries were straight (Fig. 2A and table S1). These results were confirmed by optical tweezer experiments. When individual spiral PDF machineries were stretched to four times their original lengths by optical laser trapping, they returned to their original sizes upon release (Fig. 2B; Movies S1 and S2). However, the plasticity of the spiral was lost after several stretches. Similar results were obtained for many spiral PDF machineries (table S2). In contrast, dynamin-released straight PDF

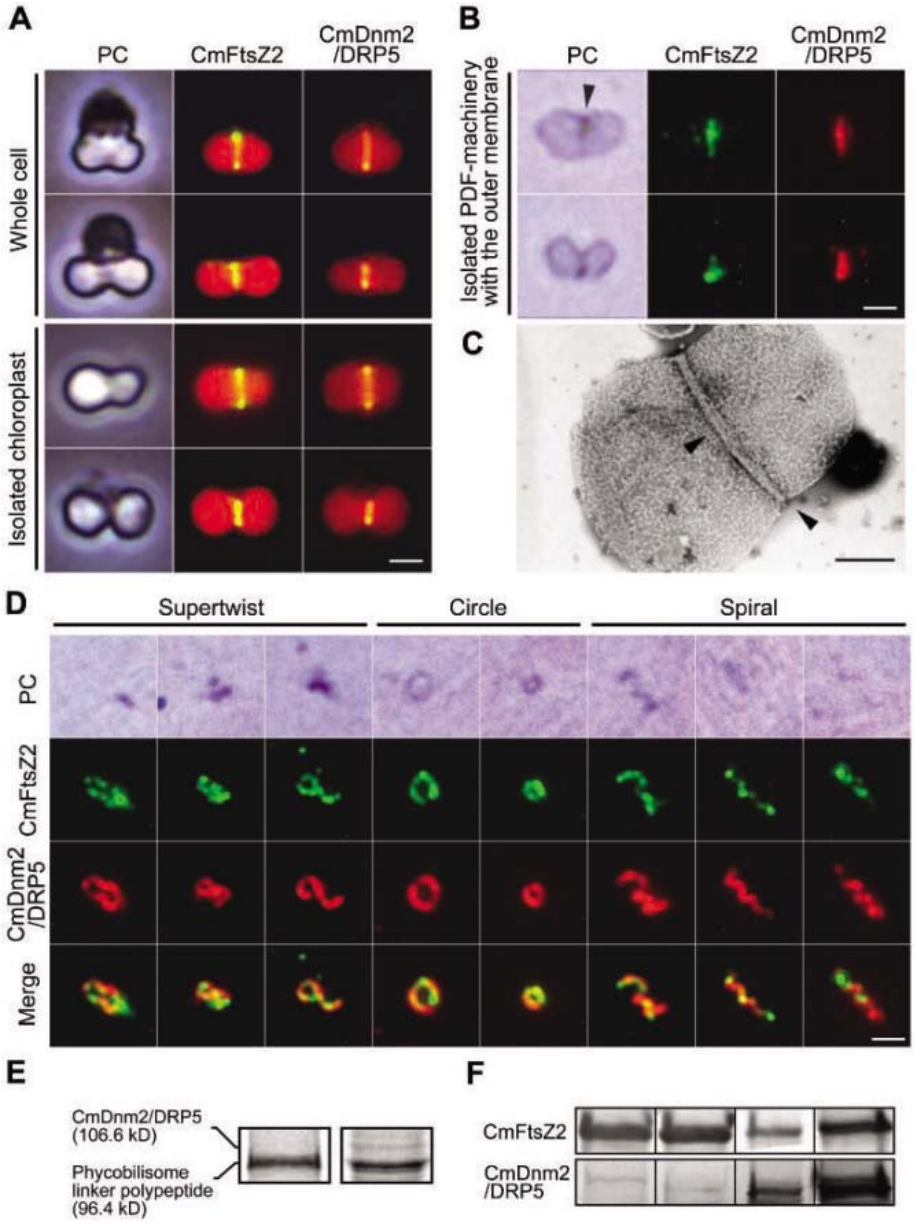


**Fig. 1.** Isolation of PDF machineries from dividing chloroplasts in *C. merolae* cells. (**A**) Phase-contrast and immunofluorescence images of the FtsZ (CmFtsZ2; yellow-green) and dynamin (CmDnm2/DRP5; orange) rings of whole cells and isolated chloroplasts at the early and late phases of chloroplast division. (**B**) Phase-contrast and immunofluorescence images of the PD (arrowhead), FtsZ, and dynamin rings of isolated PDF machineries with the outer membrane at the early and late phases of chloroplast division. (**C**) Electron micrograph of an isolated PDF machinery (arrowheads) with the outer membrane. (**D**) Phase-contrast and immunofluorescence images of the PD, FtsZ, and dynamin rings and merged images of isolated intact PDF-machineries showing supertwisted, circular, and spiral structures. (**E**) SDS-PAGE of isolated PDF machineries with the outer membrane (left) and isolated PDF-machinery fractions (right). (**F**) Immunoblotting analysis of CmFtsZ2 and CmDnm2/DRP5 proteins at each isolation step: (from left to right) whole cell, isolated chloroplast, isolated PDF machinery with the outer membrane, and isolated PDF-machinery fraction. Scale bars: 1 μm (A, B, D), 500 nm (C).

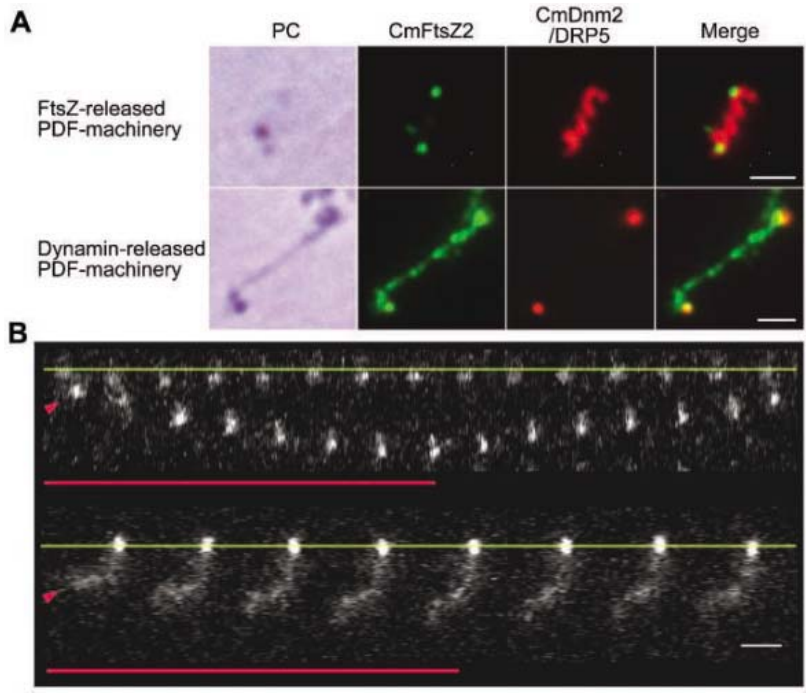


**Fig. 2.** Immunofluorescence images and manipulation of FtsZ- or dynamin-released PDF machineries. (**A**) Fluorescence images of FtsZ- (top) and dynamin- (bottom) released PDF machineries including the PD (phase-contrast, PC), FtsZ (CmFtsZ2; green), and dynamin (CmDnm2/DRP5; orange) rings and merged images. (**B**) Manipulation of an intact PDF machinery (top) and a dynamin-released PDF machinery (bottom) with optical tweezers. The base line of one end of each of the PDF machineries is fixed to the cover glass (yellow lines), while the other end is trapped by the optical tweezers (arrowheads) and an infrared laser (red lines). Scale bar, 1 μm.

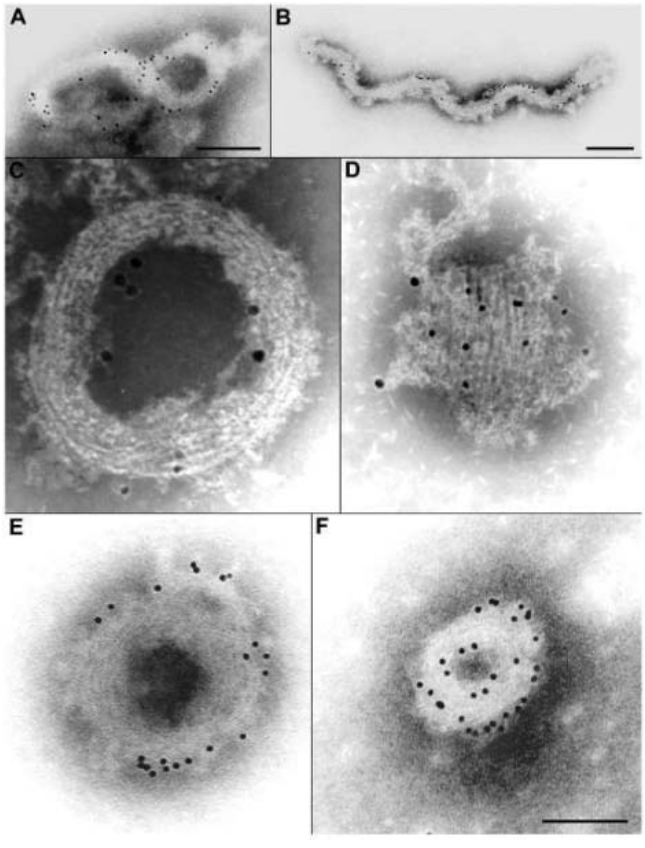


**Fig. 3.** Immunoelectron micrographs showing the distributions of dynamin and FtsZ proteins in isolated PDF machineries after negative staining. (**A** and **B**) Immunogold particles (indicating dynamin) form a line along the supertwisted (A) and spiral (B) PDF machineries at the early phase of chloroplast division. (**C** to **F**) Circular images of PDF machineries [(C), (E), and (F)] and a side image (D) at the late phase of chloroplast division. Large (indicating FtsZ) and small (indicating dynamin) immunogold particles are localized on the inside and outside of the filamentous PDF machineries, respectively [(C) and (D)]. Most of the immunogold particles (indicating dynamin) are distributed on the outside of the PDF machineries [(C) and (E)], as well as in the loosened region (C) and side view (D), and are localized between thin filaments. Upon contraction, the immunogold particles move from the outer peripheral area [(C) to (E)] to the inside (F) of the PDF machinery. Scale bars: 200 nm [(A) and (B)], 100 nm [(C) to (F)].

machineries were unable to recover from stretch (Fig. 2B and table S2). Thus, dynamin generates the motive force for contraction.

To reveal the dynamics of the PD, dynamin, and FtsZ rings in the PDF machineries, we examined isolated PDF machineries by immunoelectron microscopy. Immunogold particles indicating dynamin signals were located in a spiral line along the periphery of the supertwisted (Fig. 3A) and spiral (Fig. 3B) PDF machineries. However, the dynamin-released region became loose (fig. S5). After treatment with OG for 20 min, the disassembled PDF machineries included a smooth PD ring as a bundle of fine filaments with dynamin colocalized between the filaments (fig. S5), supporting the notion that the dynamin ring generates the motive force for contraction with the PD ring. As the contraction of the PDF machineries progressed, the supertwisted (spiral) PDF machineries changed into compact circles (Fig. 3, C to F). Immunogold particles indicating the FtsZ signals were located on the inside of the filamentous circular PD ring where the membrane was dissolved (Fig. 3C). In chloroplasts in vivo, these rings seemed to be linked with each other through holes that appeared on the groove of the division site, as revealed by scanning electron microscopy (fig. S6). In contrast, the immunogold particles indicating dynamin signals were found on the outside of the filamentous circular PD ring (Fig. 3, C to E; fig. S5), and some of the particles could be seen between the thin filaments (Fig. 3, C and D; fig. S5). When the contraction occurred during the late phase of chloroplast division, the dynamin signals moved from the outside to the inside of the constricted PDF machineries (Fig. 3, E and F). It is thought that the dynamin molecules finally pinch off the membranes in the bridge between daughter chloroplasts.

The existence of membrane-free PDF machineries suggests that there is a linking structure through the membrane between the inner (FtsZ and inner PD) and outer (outer PD and dynamin) rings. These linkers must differ from MinD, MinE, and ARC6, which are involved in the formation of the FtsZ ring in plants (*12*), because these genes were not found in the complete *C. merolae* genome (*13*). We suggest that the function of dynamin in the operation of PDF machineries can be explained by two steps: first, as a mediator of filament sliding at the early phase of chloroplast division, and then as a pinchase to pinch off the neck of the dividing chloroplast at the late phase (fig. S7). Just before contraction of the dividing chloroplast, the dynamin vesicles would move from the cytosol to the outside of the outer PD ring and release dynamin to form a PDF machinery with the PD ring, which is a bundle of fine filaments 5 to 7 nm in diameter (*2*, *3*). It has been thought that contraction of the PD ring through sliding of the PD-ring fine filaments is caused by myosin-like proteins (*2*), but no genes encoding myosin or myosin-like proteins were found in the *C. merolae* genome (*13*). Thus, it is likely that dynamin, rather than myosin-like

proteins, drives the sliding of the PD-ring fine filaments and causes the contraction required for chloroplast division (fig. S7). Dynamin was originally identified as a nucleotide-dependent microtubule-binding protein from calf brains (*14*). There, GTPase dynamin molecules in the PDF machinery appeared to function as cross-bridges that underwent microscopic movements to drive the sliding of the microtubule filaments in the PD ring during the early phase of chloroplast division (figs. S5 and S7).

At the late phase of chloroplast division, the dynamin molecules move from the surface of the contracted PDF machinery to the inside of the machinery (fig. S7). When the dynamin molecules are associated with the membrane at the final stage of chloroplast division and interact with the lipid membranes, they would act as a pinchase to pinch off the membranes at the bridge between the daughter chloroplasts (fig. S7).

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### Supporting Online Material

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# Human IRGM Induces Autophagy to Eliminate Intracellular Mycobacteria


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**Immunity-related p47 guanosine triphosphatases (IRG) play a role in defense against intracellular pathogens. We found that the murine Irgm1 (LRG-47) guanosine triphosphatase induced autophagy and generated large autolysosomal organelles as a mechanism for the elimination of intracellular *Mycobacterium tuberculosis*. We also identified a function for a human IRG protein in the control of intracellular pathogens and report that the human Irgm1 ortholog, IRGM, plays a role in autophagy and in the reduction of intracellular bacillary load.**


The immunity-related guanosine triphosphatases (GTPases) or IRGs (*1*), also known as p47 GTPases (*2*), play a role in innate immunity against intracellular pathogens (*2*, *3*). There are 23 complete *Irg* genes in the mouse genome (*1*), whereas only 3 identifiable *IRG* genes are seen in the human genome (fig. S1). Murine IRG GTPases play a role in the control of intracellular pathogens (*1*, *2*, *4*, *5*), but the functional role, if any, of human IRGs is unclear. Furthermore, most murine *Irg* genes are inducible by interferon-γ (IFN-γ), whereas the human *IRG* genes are not (*1*). In mice, IIGP1 (Irga6) (*5*) affects the integrity of vacuoles in *Toxoplasma*, whereas LRG-47 (Irgm1), acting in defense against intracellular *Myco-*


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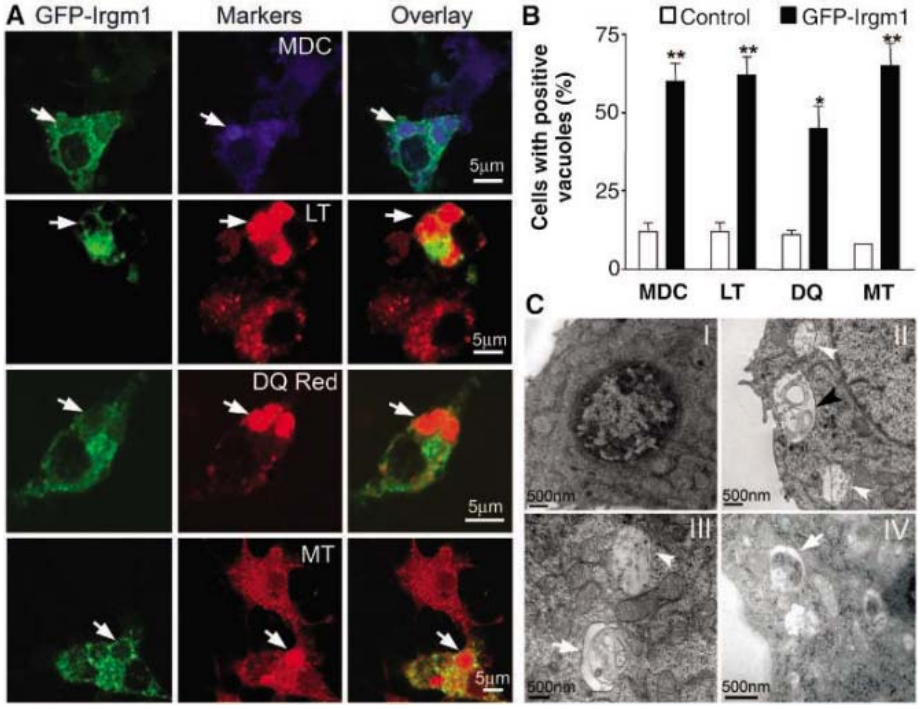


**Fig. 1.** Irgm1 induces autophagic organelles in macrophages. (**A**) RAW 264.7 cells were transiently transfected with GFP-Irgm1 and labeled for a series of markers: 50 μM MDC for 30 min, 100 nM LT for 2 hours, and 10 μg/ml of DQ Red BSA or 500 nM MT for 30 min. Arrows indicate Irgm1-induced vacuoles that were absent in the control cells. (**B**) Quantification of cells with autophagic organelles that were positive for markers tested. Results are shown as the means (% of cells with marker positive vacuoles) ± SEM; $*P < 0.05$, $**P < 0.01$ [analysis of variance (ANOVA)]. (**C**) RAW 264.7 cells were mock transfected and treated without (I) or with (II) IFN-γ. The IFN-γ–treated cells with autolysosomes containing degraded material (white arrowheads) or with double-membrane profiles in the process of engulfment of target organelles (black arrowhead) are shown. (III and IV) Cells transfected with GFP-Irgm1 show both early autophagosomes (arrows) and autolysosomes (arrowhead) that resemble the vacuoles in IFN-γ–treated cells.

*bacterium tuberculosis* (*4*, *6*), may participate in IFN-γ–induced autophagy (*7*). Autophagy is a cellular homeostasis mechanism, whereby portions of the cytosol and damaged organelles (*8*) or intracellular pathogens and their products (*9*–*16*) are sequestered into an autophagosome for degradation in autolysosomes. We investigated the role of the murine Irgm1 GTPase and its putative human ortholog IRGM in autophagy and tested whether human IRGM plays a role in control of intracellular mycobacteria.

IFN-γ induces autophagy in macrophages (*7*). Irgm1 is a downstream effector of IFN-γ that has been implicated as a potential participant in this process. We tested the effects of Irgm1 in the absence of IFN-γ stimulation. Expression in macrophages of green fluorescent protein fused to Irgm1 (GFP-Irgm1) induced the formation of large vacuoles (>3 μm) without IFN-γ stimulation (Fig. 1). The Irgm1-induced profiles were positive for monodansylcadaverine (MDC), a preliminary screening marker for autophagic organelles (Fig. 1A). The proportion of cells with $MDC^+$ vacuoles in GFP-Irgm1–transfected cells was 60 ± 4% versus 12.1 ± 1.9% in control samples ($P < 0.01$) (Fig. 1B). The classical inhibitors of autophagy, 3-methyladenine (3MA) and wortmannin, caused a marked reduction in the fraction of Irgm1 transfected cells with $MDC^+$ vacuoles in cells (53.3 ± 10.4% in samples without inhibitors versus 19 ± 4.5% with 3MA and 21.3 ± 3.5% with wortmannin; $P < 0.01$) (fig. S2).

The Irgm1-induced vacuolar compartments were positive for the acidotropic dye LysoTracker Red (LT) (Fig. 1, A and B), indicating an acidic nature of the vacuoles. The proportion of cells transfected with Irgm1 that had $LT^+$ large vacuoles was 62 ± 4% versus 12 ± 2% of control cells ($P < 0.01$). The large Irgm1-induced vacuoles were also proteolytically active as detected by fluorophore dequenching on proteolysis of bovine serum albumin conjugated to 4,4-difluoro-4-bora-3a,4a-diaza-*s*-indacene (DQ Red BSA) (Fig. 1A). The DQ Red $BSA^+$ profiles were detected in 45 ± 5% of GFP-Irgm1–transfected cells, compared to 11 ± 1% of control cells ($P < 0.05$) (Fig. 1B). The Irgm1-induced DQ Red $BSA^+$ (Fig. 1A) vacuoles were indistinguishable from those induced by IFN-γ (fig. S3).

During formation, autophagosomes sequester cytoplasmic contents and often include mitochondria (*17*). The presence of mitochondrial material inside a vacuole is indicative of a compartment representing a bona fide autophagosome. Irgm1-induced vacuoles were positive for Mitotracker Red (MT) (Fig. 1A), with a sevenfold increase in $MT^+$ cells among GFP-Irgm1–transfected cells (65 ± 5%) compared to controls (9.4 ± 3.5%; $P < 0.01$) (Fig. 1B). These characteristics are consistent with the interpretation that Irgm1-induced vacuoles were large autolysosomes.

Irgm1-induced profiles were examined by electron microscopy. Vacuoles were not observed in mock-transfected control cells (Fig. 1C, panel I; fig. S4) but were abundant in cells treated with IFN-γ (Fig. 1C, panel II, arrowheads). These compartments contained internal membranes. Similar types of vacuoles were observed in cells that were transfected with GFP-Irgm1 (Fig. 1C, panel III, arrowhead), consistent with autolysosomal nature of Irgm1-induced organelles. We also detected double membrane structures in GFP-Irgm1 transfected cells (Fig. 1C, panels III and IV, arrows), a morphological signature of nascent autophagosomes, known as phagophores or isolation membranes (*18*). Thus, in addition to large vacuoles representing autolysosomal structures, Irgm1 induces early autophagosomal organelles.

We next examined whether Irgm1 was necessary for early stages of the autophagosomal pathway. The formation of early autophagosomal precursors and newly completed autophagosomes is quantifiable by following microtubule-associated protein light-chain 3 (LC3) changes. LC3 (Atg8) exists in two forms: the cytosolic species LC3-I with an electrophoretic mobility corresponding to the relative molecular mass ($M_r$) of 18 kD, and its membrane-associated form, LC3-II, conjugated C-terminally to phosphatidylethanolamine, with an apparent $M_r$ of 16 kD. The latter form, LC3-II, inserts into the membrane of nascent autophagosomes and correlates with the appearance of $LC3^+$ autophago-

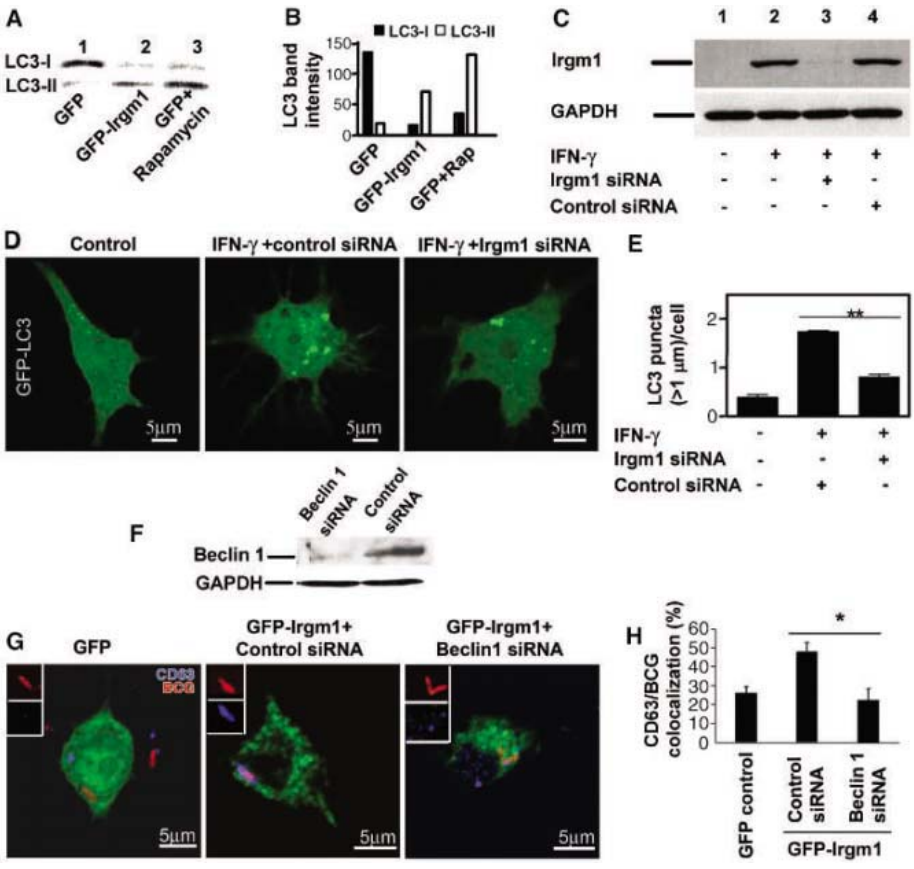


**Fig. 2.** Irgm1 is required for IFN-γ–induced autophagy and transfer of mycobacteria from immature phagosomes to compartments with late endosomal/lysosomal characteristics. (**A**) RAW 264.7 cells were transiently transfected either with GFP and incubated with or without rapamycin (50 μg/ml) for 4 hours or transfected with GFP-Irgm1. Immunoblot analysis of cells was carried out with antibody to LC3 (anti-LC3). (**B**) Quantification of the LC3 band intensity. (**C**) RAW 264.7 cells were transfected with Irgm1 siRNA or control siRNA and incubated for 24 hours with or without IFN-γ (200 U/ml). Cells were processed for immunoblotting and probed for Irgm1. (**D** and **E**) Cells were transiently cotransfected with enhanced GFP (EGFP)–LC3 and control siRNA or Irgm1 siRNA and treated with IFN-γ (200 U/ml) for 24 hours, and the number of GFP-LC3 puncta per cell was quantified. Results are shown as the means ± SEM; $^{**}P < 0.01$ (ANOVA); $n = 100$ cells from three independent experiments. (**F**) Knockdown of Beclin1 by siRNA examined by immunoblotting with anti-Beclin. (**G** and **H**) RAW 264.7 cells were transfected with GFP or cotransfected with GFP-Irgm1 and control siRNA or siRNA against Beclin 1. Cells were infected with Texas Red–labeled *M. tuberculosis* var. *bovis* BCG for 15 min and chased for 1 hour. Phagosomal maturation was assessed by colocalization of BCG phagosomes (red) with lysosomal marker CD63 (blue) with anti-CD63 followed by incubation with secondary antibody conjugated to Alexa Fluor 647. (G) Main panels show merged three-color fluorescence images, and insets show regions of interest with BCG phagosomes rendered as single-channel fluorescence (top, red; bottom, blue). Results are shown as the means ± SEM; $^{*}P < 0.05$ (ANOVA).

somes (*19*). Increasing levels of LC3-II on immunoblots can be used to document induction of autophagy (*19*). The intensity of the LC3-II band (Fig. 2, A and B) was increased in cells transfected with GFP-Irgm1 relative to the control cells transfected with GFP alone. The Irgm1-induced LC3-II levels were comparable to the effects of rapamycin, a conventional inducer of autophagy (Fig. 2, A and B). Next, we tested whether Irgm1 was responsible for IFN-γ–induced autophagy. For these studies, we reduced the expression of endogenous Irgm1 by small interfering RNA (siRNA) in IFN-γ–stimulated cells, as confirmed by immunoblotting (Fig. 2C). Once lipidated, LC3 undergoes transition from the cytosolic form to a membrane-bound form, which is then scored as cytoplasmic puncta by fluorescence microscopy (*19*). RAW 264.7 cells were cotransfected with GFP-LC3 and Irgm1 siRNA or control scrambled siRNA, and the formation of GFP-LC3 puncta was analyzed upon stimulation with IFN-γ. In cells subjected to Irgm1 inhibition, there was a decrease ($P < 0.01$) in the number of IFN-γ–induced LC3 puncta (>1 μm) (Fig. 2, D and E). Thus, Irgm1 stimulates early stages of autophagy, including the conversion of LC3-I to LC3-II, and Irgm1 is required for the autophagic pathway induced by IFN-γ.

Irgm1 controls *M. tuberculosis* infections by a process that involves enhanced mycobacterial phagosome maturation (*4*). To determine whether Irgm1-dependent mycobacterial phagosome maturation requires a functional autophagic pathway, we examined the effects of siRNA inhibition by targeting factors essential for the execution of autophagy. Beclin 1 (Atg6), a tumor suppressor that is critical to autophagy (*20*), forms a complex with the phosphatidylinositol 3-kinase hVPS34 (*21*). RAW cells were cotransfected with GFP-Irgm1 and Beclin 1 siRNA. Beclin 1 inhibition was documented by immunoblot analysis (Fig. 2F). The cells were infected with *M. tuberculosis* var. *bovis* bacille Calmette-Guérin (BCG), and phagosomal maturation was quantified by analyzing colocalization with the late endosomal marker CD63. The expression of Irgm1 caused increased maturation of BCG phagosomes. There was a significant reduction in Irgm1-induced maturation of BCG phagosomes in cells cotransfected with Beclin 1 siRNA (Fig. 2, G and H). The reduced expression of Beclin 1 and Atg7 also inhibited IFN-γ–induced BCG phagosomal maturation (fig. S5). Beclin 1 plays an essential role in autophagy but is not required for the conventional endosomal/lysosomal pathway (*22*, *23*). Thus, Irgm1 induces autophagy, whereas the Irgm1-stimulated transfer of mycobacteria to compartments with late endosomal or lysosomal characteristics depends on the autophagic pathway.

The human *IRGM* gene is syntenic with the mouse gene *lrgm1* (encoding LRG-47), and its expression at the mRNA level has been documented (*1*). We first examined IRGM expression at the protein level in human cells by immunoblotting, using siRNA inhibition to identify the protein band. U937 cells were transfected with either control siRNA or IRGM siRNA and were then probed with the antibody to IRGM. We observed a 20-kD band that was specifically down-regulated in cells treated with IRGM siRNA (Fig. 3A). We also detected an identical protein in the other human cells that were tested (293T and HeLa). The identity of the band as IRGM was established in all cells tested by siRNA inhibition (Fig. 3A). Thus, the IRGM protein is indeed expressed in human cells.

We next tested the hypothesis that human IRGM was participating in autophagy, by analogy to its mouse counterpart Irgm1. Human IFN-γ (hIFN-γ) induced autophagy in the human macrophage cell line U937 (Fig. 3B and fig. S6). IFN-γ–induced autophagy in human macrophages was dependent on a critical au-

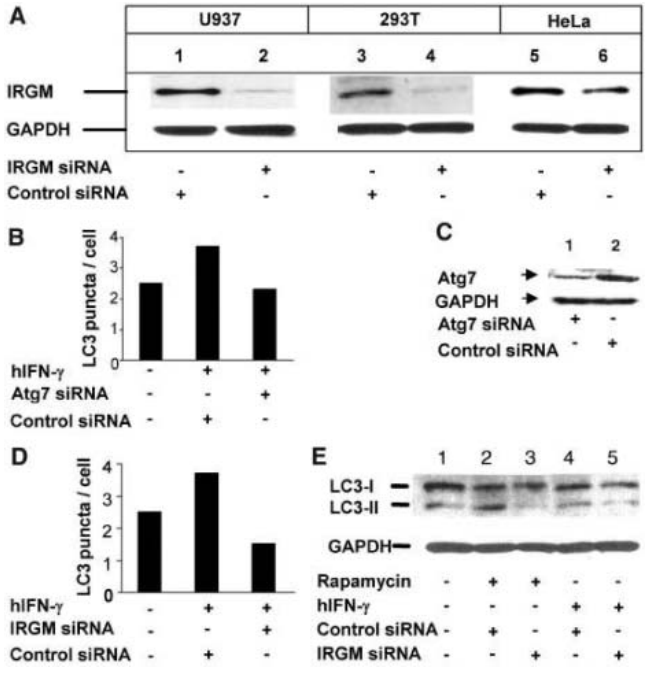


**Fig. 3.** Role of IRGM in autophagy in human cells. (**A**) Human cell lines U937, 293T, and HeLa were transfected with either control siRNA or IRGM siRNA. Cells were lysed and analyzed by Western blotting with affinity-purified anti-IRGM. IRGM siRNA, but not control siRNA, reduced the expression of IRGM in all the three cell lines. (**B**) U937 cells were transiently transfected with GFP-LC3 and control siRNA or siRNA to Atg7 and treated with hIFN-γ (300 U/ml) for 24 hours, and EGFP-LC3 puncta were quantified. (**C**) Immunoblotting showing the extent of Atg7 inhibition by siRNA corresponding to the experiment in (B). (**D**) U937 cells were transiently transfected with GFP-LC3 and control siRNA or siRNA to IRGM and treated with hIFN-γ (300 U/ml) for 24 hours; EGFP-LC3 puncta were quantified. (**E**) Cells were transfected with control siRNA or IRGM siRNA, treated with hIFN-γ for 24 hours or with rapamycin (50 μg/ml) for 4 hours, and processed for immunoblotting with anti-LC3.

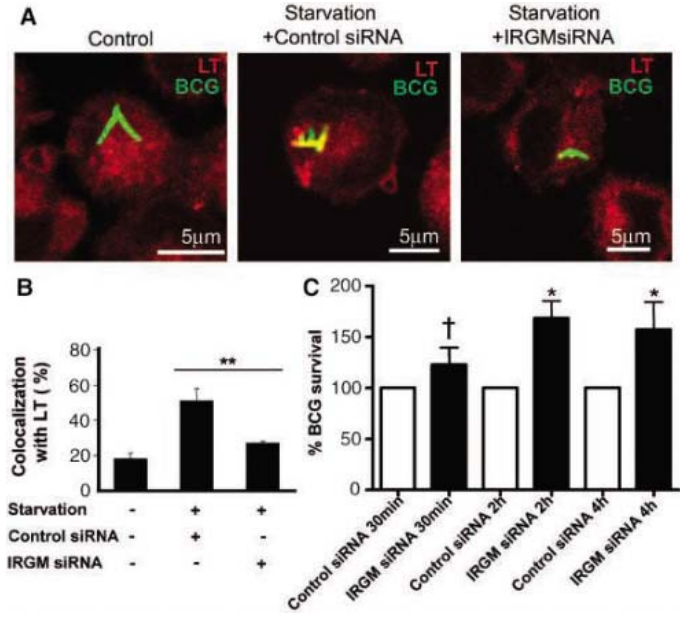


**Fig. 4.** Human IRGM promotes BCG phagosomal maturation and inhibits the survival of intracellular mycobacteria. (**A**) U937 cells were transfected with control or IRGM siRNA and infected with BCG. Cells were induced for autophagy by starvation for 2 hours. Acidified compartments (late endosomes/lysosomes) were labeled with LT. (**B**) Quantification of $LT^+$ phagosomes. Data represent the means ± SEM from three independent experiments; $^{**}P < 0.01$ (ANOVA). (**C**) U937 cells transfected with control or IRGM siRNA were infected with BCG for 1 hour, chased for 30 min, 2 hours, or 4 hours, and lysed to quantify bacterial survival by counting colony-forming units. Results are shown as the means ± SEM; †, $P > 0.05$; *, $P < 0.05$ (ANOVA).

tophagy factor, Atg7. Atg7 inhibition by siRNA (Fig. 3C) resulted in a decrease of IFN-γ–induced LC3 puncta in U937 cells (Fig. 3B and fig. S6A). In U937 cells treated with hIFN-γ and then transfected with either IRGM or control (scrambled) siRNA, the hIFN-γ–dependent increase in GFP-LC3 puncta was not affected in cells transfected with the control (scrambled) siRNA (Fig. 3D and fig. S6B). However, when cells were transfected with IRGM siRNA, the number of GFP-LC3 puncta decreased relative to that of the control cells (Fig. 3D and fig. S6B).

The role of IRGM in the conversion of endogenous LC3-I into the LC3-II form in human macrophages was confirmed by immunoblotting. IRGM inhibition by siRNA specifically reduced the amounts of LC3-II in cells treated with hIFN-γ (Fig. 3E). Moreover, when autophagy was induced with the conventional inducer rapamycin, IRGM siRNA inhibited the formation of LC3-II (Fig. 3E). Thus, IRGM is necessary for the execution of the autophagic pathway in human macrophages.

To determine whether human IRGM is involved in the resistance to mycobacteria in a manner similar to that of Irgm1 in murine cells (*4*), we examined the status of mycobacterial phagosomes. U937 cells were transfected with either control siRNA or IRGM siRNA, and autophagy was induced by starvation. Acidification of mycobacterial phagosomes was monitored by LT staining (Fig. 4, A and B). The proportion of mycobacterial phagosomes that tested positive for LT increased (50.6%) upon starvation as compared with the control cells (21%) ($P < 0.01$). In contrast, cells transfected with IRGM siRNA showed only 28% of LT colocalization with mycobacterial phagosomes upon induction of autophagy by starvation (Fig. 4, A and B). Thus, not only does IRGM participate in autophagy in human macrophages, but IRGM-dependent processes are also required for autophagy-induced BCG phagosome maturation in human cells. To test whether IRGM affects mycobacterial survival in human macrophages, we transfected human U937 cells with either control siRNA or IRGM siRNA, infected them with mycobacteria, and examined bacterial viability by counting colony-forming units in a time-course experiment after infection. Increased bacterial survival was observed in cells that were treated with IRGM siRNA compared to that in control cells (Fig. 4C). Thus, IRGM, the human ortholog of Irgm1 (LRG-47), acts as its functional equivalent in autophagy and plays a role in the control of intracellular mycobacteria in human cells.

IFN-γ induces multiple microbicidal pathways, with IRG (*2*, *3*) and autophagy (*7*) representing two of its effectors linked together in this work. The Irgm1-induced organelles represent autophagosomes and autolysosomes that are in transit through various stages of development, with terminal vacuoles that are much larger than conventional autolysosomes. Thus, Irgm1-stimulated processes allow for enhanced growth of individual autophagic organelles and may be important for efficient capture of geometrically challenging pathogens, such as clumps of *M. tuberculosis* (movie S1). Unlike the IRG family in mice, the human IRGM is not responsive to IFN-γ and is constitutively expressed (*1*). Nevertheless, IRGM participates in IFN-γ–induced or conventionally induced (by rapamycin or starvation) autophagy in human macrophages, indicating a more general role for IRGM in autophagy. IRGM participates in conferring resistance against mycobacterial infections, fulfilling a role analogous to that of Irgm1 in mice. In conclusion, this study demonstrates a role for the IRG proteins in autophagy, thereby uncovering the mechanism of action by which this class of host defense factors confers resistance to intracellular pathogens.


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**Supporting Online Material**

www.sciencemag.org/cgi/content/full/1129577/DC1
Materials and Methods
Figs. S1 to S6
References
Movie S1


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# Humanization of Yeast to Produce Complex Terminally Sialylated Glycoproteins


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**Yeast is a widely used recombinant protein expression system. We expanded its utility by engineering the yeast *Pichia pastoris* to secrete human glycoproteins with fully complex terminally sialylated N-glycans. After the knockout of four genes to eliminate yeast-specific glycosylation, we introduced 14 heterologous genes, allowing us to replicate the sequential steps of human glycosylation. The reported cell lines produce complex glycoproteins with greater than 90% terminal sialylation. Finally, to demonstrate the utility of these yeast strains, functional recombinant erythropoietin was produced.**


The half-life and therapeutic potency of most glycoproteins, with the notable exception of antibodies, is dependent on the presence of terminal sialic acid. The exposure of other terminal sugars such as mannose, *N*-acetylglucosamine, and galactose on a glycoprotein leads to clearance by sugar-specific receptors or lectins (*1*, *2*). Because most therapeutic glycoproteins require sialylation, their production to date has relied on mammalian hosts, which are able to perform humanlike N-glycosylation, including the ability to add terminal sialic acid. Yeast and filamentous fungi offer numerous advantages as recombinant protein expression systems when compared with mammalian cell culture, including higher recombinant protein titers,


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shorter fermentation times, and the ability to grow in chemically defined media. However, wild-type yeast glycosylate proteins with high-mannose type N-glycans (*3*) (Fig. 1A), which reduce half-life and compromise therapeutic function. Our laboratory has been engineering human glycosylation pathways into fungal hosts, including the yeast *P. pastoris* (*4*–*6*). The core repertoire of human glycosylation reactions (Fig. 1A) requires the sequential removal of mannose by two distinct mannosidases (i.e., α-1,2-mannosidase and mannosidase II), the addition of *N*-acetylglucosamine (by *N*-acetylglucosaminyltransferase I and II), the addition of galactose (by β-1,4-galactosyltransferase), and finally the addition of sialic acid by sialyltransferase. Sialylation, the final step of human glycosylation, is particularly difficult to accomplish in yeast, because wild-type yeast lacks all four prerequisites: (i) the ability to produce the N-glycosylated precursors terminating in β-1,4-galactose, (ii) the biosynthetic capability to produce the sugar nucleotide precursor cytidine monophosphate (CMP)–sialic acid [specifically, CMP-*N*-acetylneuraminic acid (CMP-NANA)], (iii) the transporter to shuttle CMP–sialic acid into the Golgi, and (iv) a sialyltransferase to transfer sialic acid to terminal galactose on the nascent glycoprotein (fig. S1) (*7*). All of these elements must work at high efficiency to allow for the production of sialylated glycoproteins, and organelle-specific targeting of several elements is required to permit these functions to occur in concert.

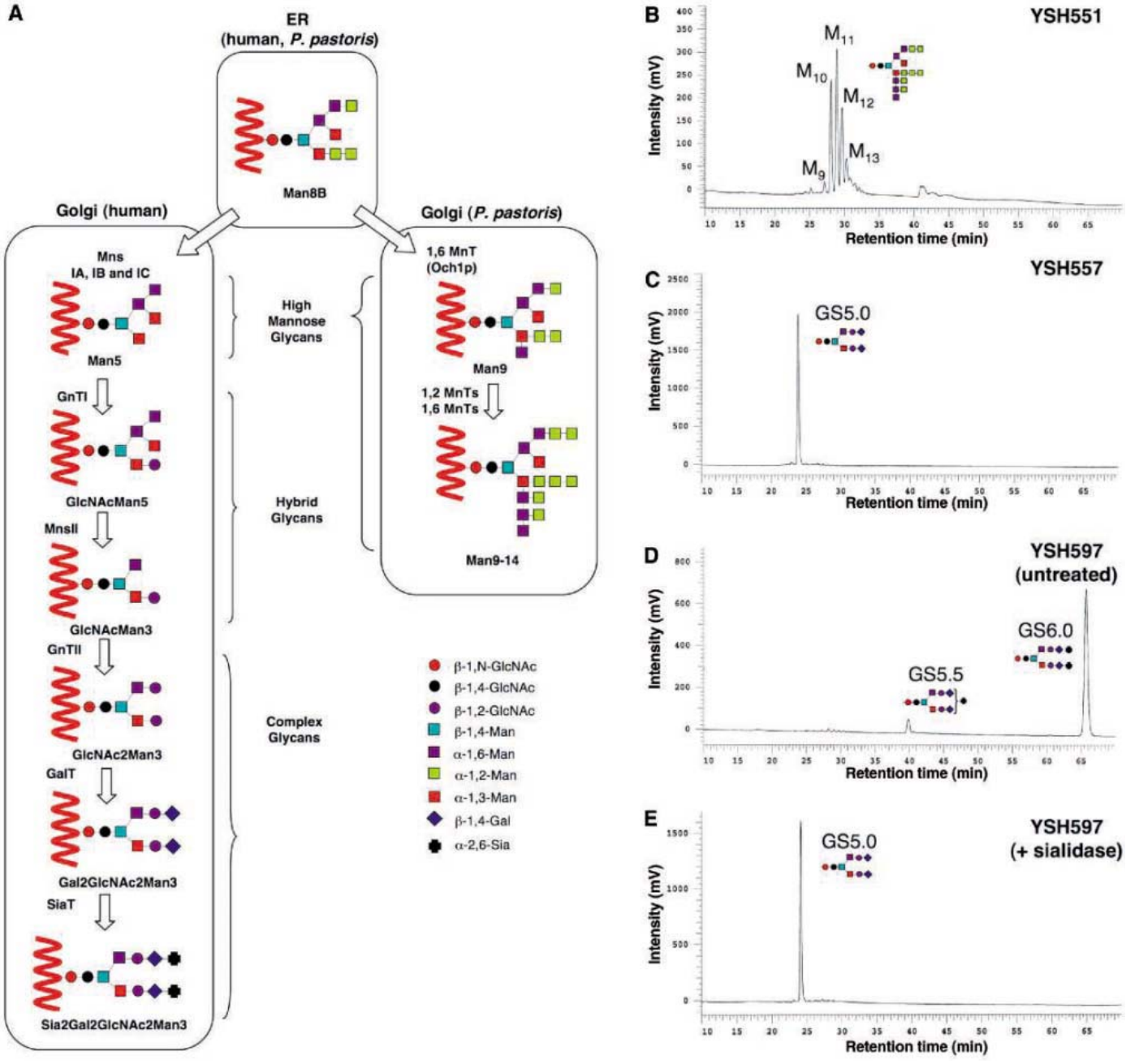


**Fig. 1.** N-linked glycosylation pathways and characterization of N-linked glycans released from recombinant rEPO. (**A**) Representative N-linked glycosylation pathways in humans and *P. pastoris*. Mns: α 1,2- mannosidase; MnsII: mannosidase II; GnTI: β 1,2-*N*-acetylglucosaminyltransferase I; GnTII: β 1,2-*N*-acetylglucosaminyltransferase II; GalT: β 1,4-galactosyltransferase; SiaT: α 2,6-sialyltransferase; MnT: mannosyltransferase. (**B** to **D**) rEPO secreted from *P. pastoris* strains YSH551 (B), YSH557 (C), and YSH597 (D) was purified from culture supernatants by Ni-affinity chromatography. Glycans were released by PNGase-F treatment and labeled with 2-AB before HPLC analysis. (**E**) Glycans secreted from YSH597 containing sialic acid were treated with α-2,3-/-2,6-/-2,8-sialidase. Elution times for commercial glycan standards corresponding to GS5.0, 5.5, and 6.0 were 24, 40, and 66 min, respectively.

Erythropoietin (EPO) is a hematopoietic glycoprotein that stimulates the differentiation of late erythroid progenitor cells to mature red blood cells (*8*). Since a recombinant source of the protein has become available, EPO has found wide therapeutic use in the treatment of anemia. EPO is a heavily glycosylated protein, with three N-glycosylation sites and up to 40% of the molecular mass attributable to its glycans (*9*, *10*). As with many glycoproteins, the therapeutic efficacy and receptor affinity of EPO relies on the degree and composition of N-glycosylation (*11*).

A previously reported glycoengineered strain of *P. pastoris* produces terminally galactosylated biantennary glycans of the complex type $Gal_2GlcNAc_2Man_3GlcNAc_2$ (i.e., Glycan Structure 5.0, GS5.0) (*12*). This strain RDP750 [Δ*och1*, Δ*pno1*, Δ*mnn4B*, Δ*bmt2*, Δ*his1*, *Kluyveromyces lactis* and *Mus musculus* uridine diphosphate (UDP)–GlcNAc transporters, *Mus musculus* α-1,2-MnsI, *Homo sapiens* β-1,2-GlcNAc transferase I, *Rattus norvegicus* β-1,2-GlcNAc transferase II, *Drosophila melanogaster* MnsII, *Schizosaccharomyces pombe* Gal epimerase, *D. melanogaster* UDP-Gal transporter, and *H. sapiens* β-1,4-galactosyltransferase] was transformed with an expression plasmid encoding for rat EPO (rEPO) to generate strains RDP762 and YSH557 (*7*). Secreted rEPO from YSH557 consisted predominantly of GS5.0 N-glycans (Fig. 1C). For comparison, the same construct was used to transform wild-type *P. pastoris* NRRL-Y11430, resulting in strain YSH551, which secreted rEPO with mostly high-mannose N-glycans that are typical for this yeast (*3*) (Fig. 1B). Both strains displayed similar growth characteristics and expressed rEPO at about the same level (~20 mg/liter), although the proteins differed significantly with respect to their N-glycosylation (Fig. 1, B and C).

To accomplish the final step of human glycosylation, the addition of terminal sialic acid, we transformed *P. pastoris* strain RDP762 with a range of DNA constructs encoding for enzymes involved in CMP–sialic acid biosynthesis, CMP–sialic acid transport, and sialic acid transfer to the nascent glycoprotein. In total, five enzymes were selected: *H. sapiens* UDP-*N*-acetylglucosamine-2-epimerase/*N*-acetylmannosamine kinase (GNE), *H. sapiens* *N*-acetylneuraminate-9-phosphate synthase (SPS), *H. sapiens* CMP–sialic acid synthase (CSS), *M. musculus* CMP–sialic acid transporter (CST), and a library of chimeric sialyltransferases linked to yeast type-II transmembrane localization peptides (ST).

More than 120 permutations of alternative CMP–sialic acid pathways, CSTs, and STs were screened. From this screen, we identified a small number of combinations displaying significant sialyltransferase activity and producing predominantly complex glycan structures. However, taking this approach we were unable to obtain glycan compositions containing >60% GS6.0 ($Sia_2Gal_2GlcNAc_2Man_3GlcNAc_2$).

To further improve the efficiency of sialylation, we codon-optimized GNE, SPS, CSS, and CST, and screened additional ST/leader fusions. The catalytic domain of mouse α-2,6-ST, fused to the *Saccharomyces cerevisiae* mannosyltransferase 1 (Mnt1) targeting signal was particularly effective. To consolidate these efforts, we cloned all five genes into a single expression vector, pSH926 (*7*). Transformation of this vector into RDP762 complemented the histidine auxotrophy of the host, while targeting the gene cluster to the *TRP2* locus of the *Pichia* genome. The resulting strain, designated YSH597, was cultured in shake flasks to secrete rEPO. Analysis of the N-glycans isolated from rEPO displayed a glycan composition that consisted predominantly of sialylated glycan structures GS6.0 (90.5%) and GS5.5 (7.9%, $SiaGal_2GlcNAc_2Man_3GlcNAc_2$) (Fig. 1D). Subsequent treatment of this sample with sialidase showed quantitative conversion to GS5.0 (Fig. 1E), confirming that GS6.0 and GS5.5 were terminally sialylated glycans.

To assess the activity of these two different rEPO glycoforms in vivo, we purified material from wild-type *P. pastoris* (YSH551) and YSH597 (*7*). The purified protein was characterized by sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) (Fig. 2A), and rEPO from wild-type *P. pastoris* showed extensive heterogeneity consistent with hyperglycosylation and the range of high-mannose structures found by high-performance liquid chromatography (HPLC) (Fig. 1B). In contrast, rEPO from YSH597 showed a more uniform migration pattern, consistent with the glycan uniformity found by HPLC (Fig. 1D). As expected, when both samples were treated with peptide N-glycosidase F (PNGase-F), to remove the N-glycans, the mass and uniformity of the deglycosylated material appeared similar (Fig. 2A). To compare the functionality of these two vastly different glycoforms, animal studies were performed to determine their respective erythropoietic function. As expected, rEPO produced in wild-type yeast had no measurable erythropoietic function, whereas rEPO produced in YSH597 showed a dose-dependent response consistent with a biologically active form of the protein (Fig. 2B).

We report the generation of yeast cell lines of *P. pastoris* with a substantially reengineered secretory pathway. These cell lines secrete terminally sialylated, complex, bi-antennary glycoproteins as exemplified by rEPO, as well as other recombinant proteins tested. The availability of such yeast cell lines may eliminate the need for mammalian cell culture in the future and allow for the production of therapeutic glycoproteins in a nonmammalian host. While significantly reducing production time and viral containment issues, this will also provide improvements in product uniformity and overall production economics. Previously, a panel of glycoengineered yeast cell lines displaying a limited repertoire of human glycosylation reactions allowed us to elucidate glycosylation-dependent structure activity relationships (*12*). Here, we have engineered into yeast the most complex step of human N-glycosylation, terminal sialylation, which will expand our ability to conduct structure-function investigations.

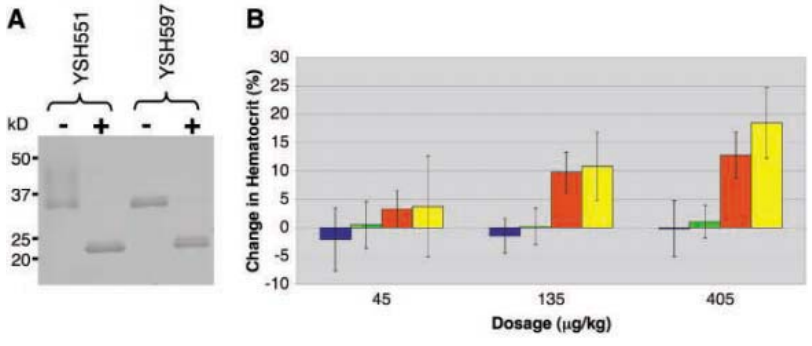


**Fig. 2.** Characterization of recombinant rEPO obtained from *P. pastoris*. (**A**) SDS-PAGE analysis of recombinant rEPO (2.5 μg, postpurification) secreted from YSH551 and YSH597 strains after incubation in the presence (+) or absence (–) of PNGase-F. (**B**) Comparative hematocrit analysis of recombinant rEPO secreted from YSH551 (blue and green bars) and YSH597 (red and yellow bars). Values correspond to days 8 (blue and red bars) and 15 (green and yellow bars) after initial injection. Data presented as mean ± SD of $n = 5$ mice per dose.

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### Supporting Online Material

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Materials and Methods
Figs. S1 and S2
References


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# An Antigen Produced by Splicing of Noncontiguous Peptides in the Reverse Order

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CD8-positive T lymphocytes recognize peptides that are usually derived from the degradation of cellular proteins and are presented by class I molecules of the major histocompatibility complex. Here we describe a human minor histocompatibility antigen created by a polymorphism in the *SP110* nuclear phosphoprotein gene. The antigenic peptide comprises two noncontiguous SP110 peptide segments spliced together in reverse order to that in which they occur in the predicted SP110 protein. The antigenic peptide could be produced in vitro by incubation of precursor peptides with highly purified 20*S* proteasomes. Cutting and splicing probably occur within the proteasome by transpeptidation.

Most nucleated cells display on their surface a broad repertoire of peptides derived from proteasome-mediated degradation of intracellular proteins and bound to major histocompatibility complex (MHC) class I molecules, which are known in humans as human leukocyte antigen (HLA) class I molecules. Surveillance of this repertoire by $CD8^+$ T lymphocytes allows the adaptive cellular immune system to detect and to eliminate cells containing foreign or abnormal proteins (*1*, *2*). The search for antigens recognized by $CD8^+$ T cells has focused on contiguous fragments of proteins expressed in malignant or infected cells. Recently, two antigenic peptides recognized by antitumor $CD8^+$ T cells were each found to be composed of the fusion of peptide fragments of the respective parental proteins after excision of an intervening segment (*3*, *4*). In one case, the excision and splicing reactions were shown to occur in the proteasome (*4*).

A $CD8^+$ cytolytic T-lymphocyte (CTL) clone, termed DRN-7, was isolated from a recipient of MHC-matched allogeneic hematopoietic cell transplantation (HCT) (*5*). In this setting, donor T cells recognizing minor histocompatibility (H) antigens, which are peptides presented on recipient cells and encoded by polymorphic non-MHC genes, can cause graft-versus-host (GVH) disease and graft-versus-leukemia (GVL) reactions (*6*). CTL DRN-7 was found to recognize an HLA-A*0301–restricted minor H antigen expressed by hematopoietic cells and to inhibit engraftment of $HLA\text{-}A^*0301^+$ human acute myelogenous leukemia cells in nonobese diabetic/severe combined immunodeficient (NOD/SCID) mice; these findings suggest that the minor H antigen may be a GVL target (*7*). To identify the gene that encodes this antigen, we screened a cDNA library constructed from the Epstein-Barr virus (EBV)–transformed B cells of the recipient. Plasmid DNA from this library was transfected into COS-7 cells, together with DNA encoding HLA-A*0301, and the transfectants were tested for their ability to stimulate CTL DRN-7. We identified a positive pool and screened DNA from individual colonies isolated from this pool. A cDNA, termed AH9C10, stimulated HLA-A*0301–dependent tumor necrosis factor (TNF) release from CTL DRN-7 (Fig. 1A). This cDNA corresponded to nucleotides 730 to 1376 of the transcript of gene *SP110*, which encode residues 213 to 425 of the SP110 nuclear body protein (*8*, *9*) (Fig. 1A). Testing truncated constructs of AH9C10 for their ability to stimulate CTL DRN-7 further localized the antigen-encoding region to a 60-nucleotide (nt) interval encoding amino acids 286 to 305 of SP110 (Fig. 1A).

Most human minor H antigens that have been defined result from nonsynonymous polymorphisms in the coding region of normal genes. Sequencing of the *SP110* alleles in EBV-transformed B cell lines derived from the transplant recipient

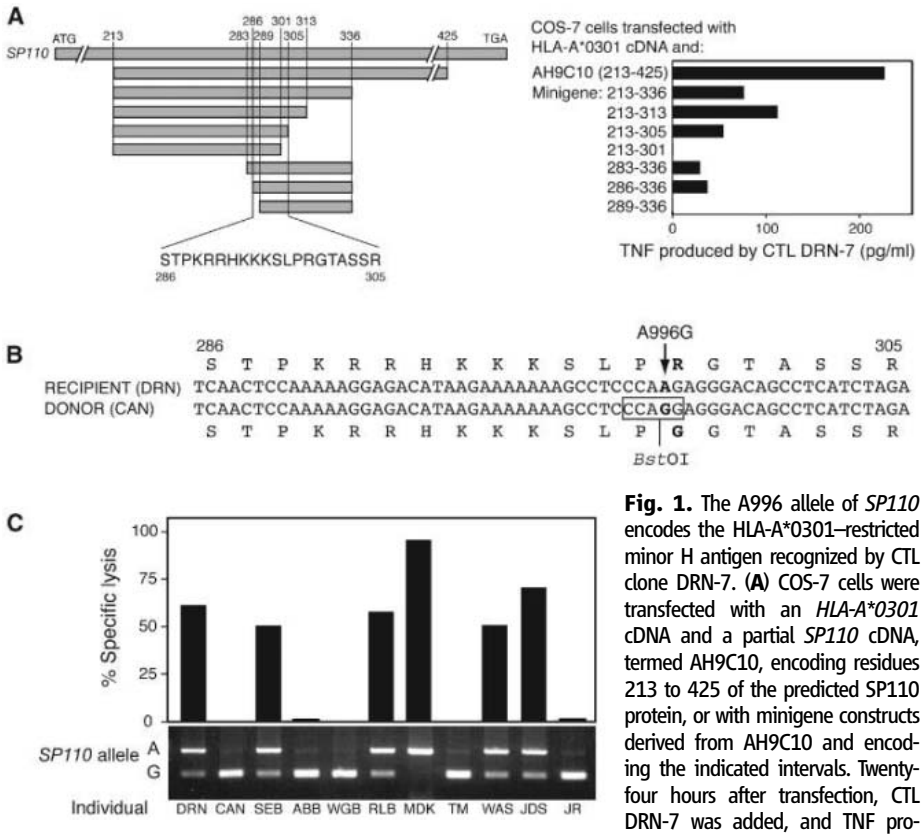


**Fig. 1.** The A996 allele of *SP110* encodes the HLA-A*0301–restricted minor H antigen recognized by CTL clone DRN-7. (**A**) COS-7 cells were transfected with an *HLA-A*0301* cDNA and a partial *SP110* cDNA, termed AH9C10, encoding residues 213 to 425 of the predicted SP110 protein, or with minigene constructs derived from AH9C10 and encoding the indicated intervals. Twenty-four hours after transfection, CTL DRN-7 was added, and TNF production was measured 24 hours later. (**B**) Nucleotide sequence of a fragment of the *SP110* alleles carried by the transplant recipient and transplant donor. The A/G polymorphism at nucleotide 996 (bold), and the corresponding amino acid polymorphism at position 299 are shown. This polymorphism is reported in databases as *rs1365776*. The Bst OI restriction site used for genotyping is indicated. (**C**) Correlation between the presence of the A996 *SP110* allele and recognition by CTL DRN-7. (Top) Lysis of $^{51}Cr$-labeled EBV–B cells from 11 representative HLA-A*0301 individuals, including the transplant recipient (DRN) and donor (CAN), by CTL DRN-7 at an effector-to-target ratio (E/T) of 10. (Bottom) Genotype of each individual at the A996G polymorphism in *SP110*.

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and donor identified an A/G polymorphism at nucleotide 996 in the *SP110* coding sequence (Fig. 1B). This polymorphism was contained within the 60-nt interval encoding the antigen recognized by CTL DRN-7 and created an amino acid substitution of Gly for Arg at position 299 (R299G) of the SP110 protein (*10*). The genotype at this polymorphism was determined in EBV–B cell lines from 64 other HLA-A*0301 individuals. A perfect correlation was observed between the presence of at least one A996 allele and susceptibility to lysis by CTL DRN-7 (Fig. 1C). In the 66 individuals tested, the frequencies of the A996 and the G996 alleles were 0.54 and 0.46, respectively.

We attempted to identify the antigenic peptide encoded by the critical 60-nt interval in the A996 allele of *SP110* by synthesizing a series of overlapping peptides that collectively spanned the predicted 20–amino acid fragment and by testing each one for recognition by CTL DRN-7 after loading onto donor EBV–B cells. None of these peptides sensitized EBV–B cells to lysis (table S1). Considering that a posttranslational modification might be required for antigenicity, we tried to enable this modification by introducing the 20–amino acid peptide STPKRRHKKKSLPRGTASSR inside donor EBV–B cells by electroporation (*4*). Cells electroporated with this peptide were strongly recognized by CTL DRN-7 (Fig. 2A), whereas cells electroporated with peptide STPKRRHKKKSLPGGTASSR, encoded by the $G^{996}$ allele, were not. The putative modification involved intracellular processing, because it was not observed when the electric shock was omitted. Moreover, in experiments where intracellular loading of the precursor peptide was obtained by prolonged incubation with very high doses of the peptide, we observed that recognition of loaded cells was dependent on the expression of transporter associated with antigen processing (TAP), which indicates a cytosolic processing step (fig. S1).

The transfection of truncated cDNA constructs had indicated that both ends of the 20–amino acid fragment were required for antigenicity (Fig. 1A). This suggested that splicing of two peptide fragments contained within STPKRRHKKKSLPRGTASSR might produce the antigen. We electroporated pairs of peptides making up nonoverlapping segments of STPKRRHKKKSLPRGTASSR into donor EBV–B cells and tested the electroporated cells for recognition by CTL DRN-7. Two decameric peptides, STPKRRHKKK (residues 286 to 295) and SLPRGTASSR (296 to 305), stimulated CTL DRN-7 when electroporated simultaneously, but not when electroporated singly (Fig. 2A). Thus, the antigenic peptide was made from the splicing of two distinct fragments. To identify the minimal fragments required, we electroporated a series of peptides truncated at either end (Fig. 2A). Removal of the N-terminal serine of either peptide abrogated recognition. Sequential removal of C-terminal residues revealed that the combination of STPK and SLPRGTAS retained the ability to stimulate CTL DRN-7. We hypothesized that the antigenic peptide was a spliced product resulting from linkage of these two fragments or parts of them. We synthesized a series of peptides containing such fragments, including peptide STPKSLPRGT, and loaded them directly onto target cells at concentrations up to 10 μg/ml. CTL DRN-7 did not recognize any of these peptides (Fig. 2B and table S2).

Peptide STPKSLPRGT did not contain a good HLA-A3–binding motif, which has leucine in position two and lysine at the C terminus (*11*, *12*). We noted that if its two constituent fragments were linked in the reverse order, the resulting peptide, SLPRGTSTPK, would contain a perfect HLA-A3–binding motif. We synthesized and loaded a peptide with this reordered sequence onto target cells, and we observed efficient recognition by CTL DRN-7, with half-maximal lysis at a peptide concentration of ~40 pM (Fig. 2B). Peptide SLPGGTSTPK, which would be derived from the G996 allele of *SP110*, was only weakly recognized at high concentrations. We tested additional reordered and spliced peptides of various lengths and observed that peptide SLPRGTSTPK was the optimal peptide, even though the longer peptides SLPRGTSTPKRR and SLPRGTSTPKR were also recognized by CTL DRN-7 at slightly higher concentrations (Fig. 2B and fig. S2).

To determine whether this spliced and reordered SP110 peptide was identical to the peptide naturally presented at the cell surface, we isolated MHC class I molecules from HLA-$A*0301^+$ EBV–B cells that were homozygous for the A996 *SP110* allele. We eluted peptides

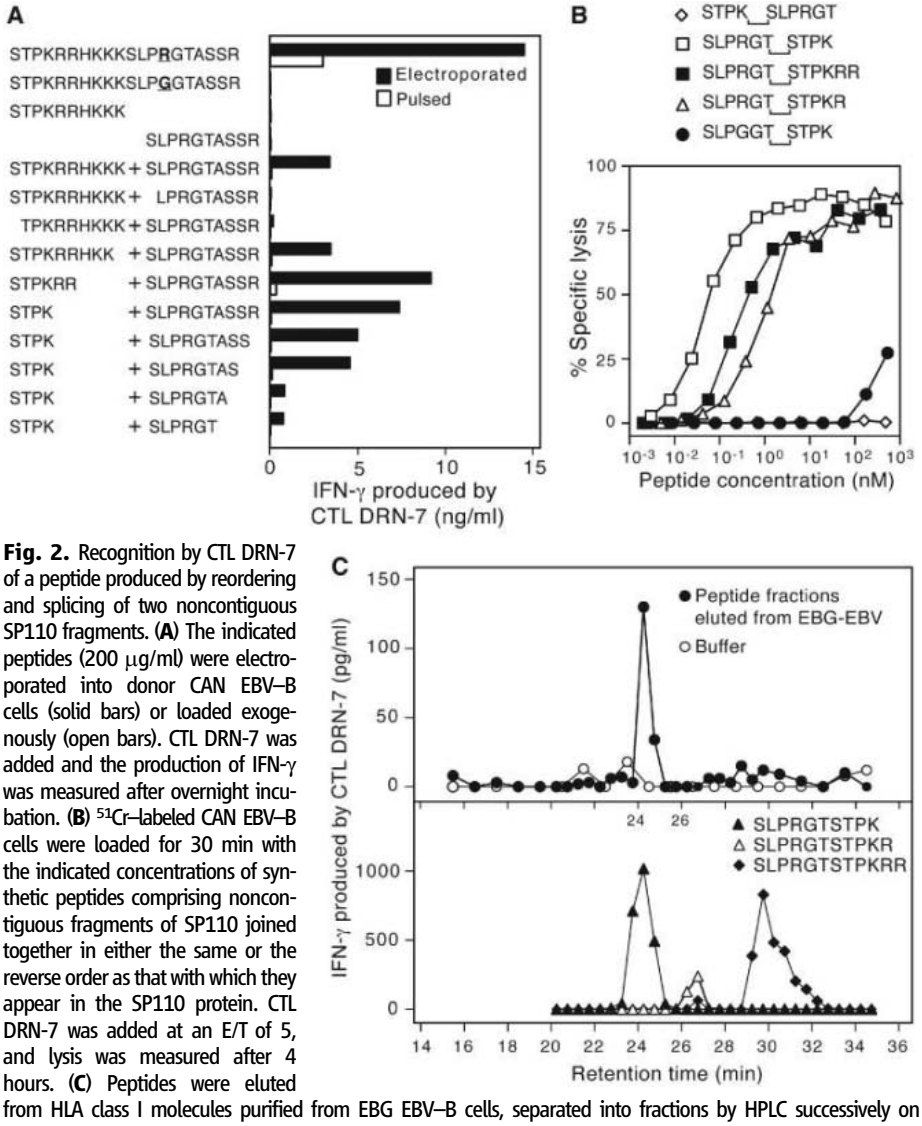


**Fig. 2.** Recognition by CTL DRN-7 of a peptide produced by reordering and splicing of two noncontiguous SP110 fragments. **(A)** The indicated peptides (200 μg/ml) were electroporated into donor CAN EBV–B cells (solid bars) or loaded exogenously (open bars). CTL DRN-7 was added and the production of IFN-γ was measured after overnight incubation. **(B)** $^{51}Cr$–labeled CAN EBV–B cells were loaded for 30 min with the indicated concentrations of synthetic peptides comprising noncontiguous fragments of SP110 joined together in either the same or the reverse order as that with which they appear in the SP110 protein. CTL DRN-7 was added at an E/T of 5, and lysis was measured after 4 hours. **(C)** Peptides were eluted from HLA class I molecules purified from EBG EBV–B cells, separated into fractions by HPLC successively on two different columns, and the fractions were tested for recognition by CTL DRN-7 (top). To rule out contamination of the HPLC system, buffer was run on the column before the eluted samples, and the fractions were tested similarly. Synthetic peptides SLPRGTSTPK, SLPRGTSTPKR, and SLPRGTSTPKRR (60 pmol each), were injected under the same HPLC conditions, and the fractions were tested for CTL recognition (bottom).

with acid and fractionated them by high-performance liquid chromatography (HPLC) on a C18 column. We identified a fraction that was able to stimulate interferon-γ (IFN-γ) release by CTL DRN-7 when loaded onto target cells. When synthetic peptide SLPRGTSTPK was chromatographed under the same HPLC conditions, the same fraction stimulated CTL DRN-7. However, we observed that peptides SLPRGTSTPK, SLPRGTSTPKR, and SLPRGTSTPKRR had similar retention times on the C18 column. We then reinjected the positive fraction obtained with the eluates into a porous graphitic carbon column, which better separates such highly hydrophilic peptides. Once again, a fraction was identified that stimulated CTL DRN-7 (Fig. 2C). When synthetic peptide SLPRGTSTPK was chromatographed on this column, the same fraction stimulated CTL DRN-7 (Fig. 2C). This was not the case with synthetic peptides SLPRGTSTPKR or SLPRGTSTPKRR. Thus, synthetic peptide SLPRGTSTPK corresponded to the peptide naturally presented to CTL DRN-7.

Previous work has identified the role of the proteasome in the production of a spliced antigenic peptide (*4*). Pretreatment of donor EBV–B cells with the irreversible proteasome inhibitor lactacystin before electroporation of STPKRRHKKKSLPRGTASSR inhibited their ability to stimulate CTL DRN-7 (fig. S3). To directly evaluate the ability of the proteasome to produce the SP110 peptide, we incubated purified 20*S* proteasomes with peptide STPKRRHKKKSLPRGTASSR and examined recognition by CTL DRN-7 of target cells loaded with the digests. Digests obtained after 1 or 2 hours of incubation were strongly recognized by CTL DRN-7, which indicated that the antigenic peptide had been produced in vitro (Fig. 3A). This was not the case when the incubation was performed in the presence of lactacystin, or with peptide STPKRRHKKKSLPGGTASSR, which is encoded by the G996 *SP110* allele (Fig. 3A).

We used HPLC coupled to mass spectrometry (MS) to identify the fragments present in the digests (Fig. 3B). This analysis revealed at least eight cleavages in the peptide, including the three cleavages required to liberate fragments STPK and SLPRGT and to allow their splicing to produce the antigenic peptide SLPRGTSTPK. This peptide was not detected by HPLC-MS in those digests. However, by using HPLC combined with tandem mass spectrometry (MS/MS), we identified the antigenic peptide SLPRGTSTPK in a digest obtained by incubation of proteasomes with peptides STPK and SLPRGTASSR. This digest was also strongly recognized by CTL DRN-7 (Fig. 3A). The antigenic peptide was detected as a doubly charged ion with *m/z* 522, whose retention time and fragmentation pattern were identical to those of the corresponding synthetic peptide (Fig. 3C). Thus, the proteasome can perform the splicing reaction required to produce antigenic peptide SLPRGTSTPK.

Our prior study of a spliced peptide indicated that splicing occurred inside the proteasome by transpeptidation involving an acyl-enzyme intermediate (*4*). This intermediate, which is transiently formed on the hydroxyl group of the side chain of the catalytic threonine, is rapidly hydrolyzed during proteolysis. In the splicing reaction, the N-terminal group of the other peptide fragment competes with water molecules to perform a nucleophilic attack of the ester bond of the intermediate, which results in a transpeptidation reaction producing the spliced peptide. In theory, this mechanism could also allow for a reordering of the peptide fragments before their ligation (Fig. 4A). The acyl-enzyme intermediate would involve fragment SLPRGT; fragment STPK would be first liberated by hydrolysis and would attack the ester bond of the intermediate with its amino group. If this model accounts for the production of the SP110 peptide, the energy required to create the new peptide bond of the spliced peptide should be recovered, through the ester bond of the intermediate, from the energy liberated by the cleavage between $Thr^{301}$ and $Ala^{302}$. A proteasomal digest of peptides STPK and SLPRGT, which correspond to the final fragments of the spliced peptide and would not need any additional cleavage, failed to produce the antigenic peptide, which confirmed the need to recover the energy of a cleaved bond (Fig. 4B). In contrast, a digest involving STPK and SLPRGTASSR did produce the antigen, whereas a digest of STPKRR and SLPRGT did not (Fig. 4B). Thus, cleavage of the bond between $Thr^{301}$ and $Ala^{302}$ is necessary to pro-

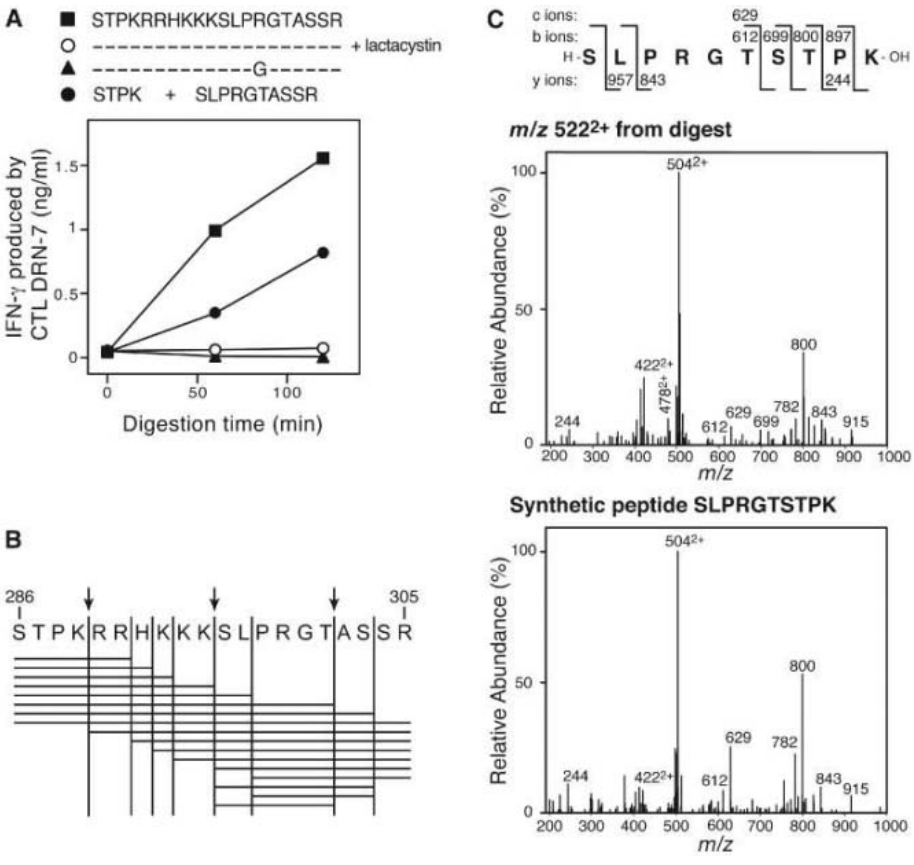


**Fig. 3.** Production of the antigenic peptide SLPRGTSTPK by the proteasome. (**A**) Recognition by CTL DRN-7 of digests obtained by incubating the indicated peptides with purified 20*S* proteasomes. The digests collected at the indicated time points were loaded onto CAN EBV–B cells and tested for recognition by CTL DRN-7. (**B**) Peptide fragments detected by MS after incubation of precursor peptide STPKRRHKKKSLPRGTASSR with 20*S* proteasomes for 60 min. Cleavage sites are indicated by vertical lines and those that are relevant for the production of the antigenic peptide, by arrows. The experimental conditions did not allow fragment quantification or the detection of small fragments (below four to five residues). A similar fragmentation pattern was observed at various digestion times. (**C**) MS/MS fragmentation spectrum of the doubly charged ion with *m/z* $522^{2+}$ observed in the digest obtained after a 180-min incubation of 20*S* proteasomes with peptides STPK and SLPRGTASSR (top), and fragmentation spectrum of the doubly charged ion (*m/z* $522^{2+}$) of the synthetic decamer SLPRGTSTPK (bottom). The fragments that were detected are indicated above the peptide sequence for N-terminal b or c ions and below for C-terminal y ions. Ion with *m/z* $504^{2+}$ is a doubly dehydrated derivative of ion $522^{2+}$.

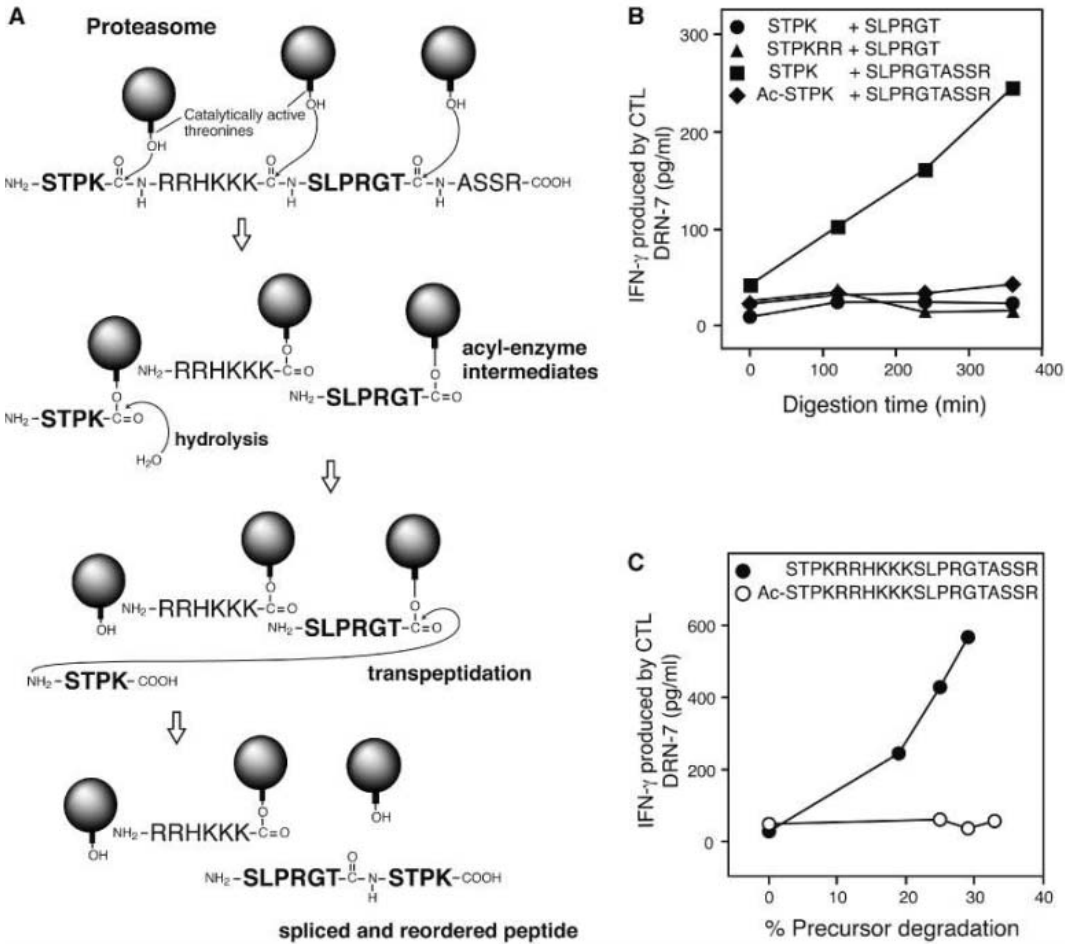


**Fig. 4.** Mechanism of peptide splicing. (**A**) Model of the splicing reaction inside the proteasome. The balls represent the catalytically active β-subunits of the proteasome with the hydroxyl group of the side chain of the N-terminal threonine. (**B**) Various synthetic peptides were combined in a pairwise manner and incubated with 20*S* proteasomes. Digests were tested for recognition by CTL DRN-7. Ac-STPK, N-α-acetylated peptide STPK. (**C**) The indicated precursor peptide or its N-α-acetylated derivative were incubated with 20*S* proteasomes. Digests were tested for recognition by CTL DRN-7. Results are expressed as a function of the percentage of degradation of the precursor peptide, as measured by HPLC-MS. Digestion times were 0, 120, 240, and 360 min.

vide the energy required to create the new bond of the spliced peptide. Acetylation of the N-terminal group of STPK prevented its ability to produce the antigenic peptide when digested with SLPRGTASSR (Fig. 4B). Similarly, acetylation of the N terminus of the single peptide STPKRRHKKKSLPRGTASSR abolished its ability to produce the antigenic peptide (Fig. 4C). Thus, consistent with our model, a free N-terminal group is required on fragment STPK to perform the nucleophilic attack of the acyl-enzyme intermediate.

Our results indicate that the reordered spliced peptide SLPRGTSTPK derived by proteasomal processing of the $Arg^{299}$ SP110 protein is the naturally processed antigen recognized by CTL DRN-7 (supporting online text) and that the splicing reaction occurs in the proteasome by transpeptidation involving an acyl-enzyme intermediate. In contrast to the two previous examples of spliced peptides (*3*, *4*), the peptide recognized by CTL DRN-7 is produced by ligation of two noncontiguous peptide fragments in the reverse order. The observation that this antigen is expressed in some normal cells (*5*) indicates that peptide splicing is not restricted to tumor cells. The proteasome, by virtue of its proteolytic capacity, participates in the generation of active transcription factor domains from inactive precursors (*13*–*17*), controls the levels of numerous regulatory proteins, and serves as the major source of peptides recognized by $CD8^+$ T cells. The ability of the proteasome to splice together peptide fragments from a protein in either the initial or reverse order has profound implications for the diversity of peptides that can be presented on the cell surface for recognition by $CD8^+$ T cells and could also have other unanticipated consequences.

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### Supporting Online Material

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# Gene Transposition as a Cause of Hybrid Sterility in *Drosophila*

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We describe reproductive isolation caused by a gene transposition. In certain *Drosophila melanogaster–D. simulans* hybrids, hybrid male sterility is caused by the lack of a single-copy gene essential for male fertility, *JYAlpha*. This gene is located on the fourth chromosome of *D. melanogaster* but on the third chromosome of *D. simulans*. Genomic and molecular analyses show that *JYAlpha* transposed to the third chromosome during the evolutionary history of the *D. simulans* lineage. Because of this transposition, a fraction of hybrids completely lack *JYAlpha* and are sterile, representing reproductive isolation without sequence evolution.

Reproductive isolation can be a by-product of divergent evolution between populations and is a necessary step in speciation. Dobzhansky and Muller described a model for the evolution of reproductive isolation wherein functional divergence between interacting loci in different lineages yields incompatible interactions in their hybrids (*1*, *2*). Although evidence for Dobzhansky-Muller interactions is well established, few genes involved in these incompatibilities have been identified and characterized (*3*). Furthermore, it remains unclear whether molecular evolutionary processes other than functional divergence cause postzygotic reproductive isolation (*2*).

The genetic basis of postzygotic isolation between *Drosophila melanogaster* and *D. simulans* has been studied previously by crossing triploid *D. melanogaster* females to heavily x-irradiated *D. simulans* males (*4*). This approach avoided the normal sterility and inviability of *D. melanogaster–D. simulans* $F_1$ hybrids, producing hybrids with backcross-like genotypes. One of these individuals, a fertile female, was used to establish a stock that carried the tiny "dot" fourth chromosome of *D. simulans* in an otherwise *D. melanogaster* genetic background. Hybrid males homozygous for the *D. simulans* fourth (*4-sim*) chromosome were completely sterile because mature sperm were immotile (*5*). Because the fourth chromosome does not recombine during meiosis (*6*), the location of the hybrid sterility gene(s) was mapped by using deletions and translocations to cytological regions 101E to 101F and/or 102A5 to 102B5 (*5*, *7*).

It remained unclear, however, whether *4-sim* hybrid male sterility was genuine or an artifact of the x-irradiation used to construct the hybrid stock. By using recently characterized mutations that rescue the viability (*8*) and fertility (*9*) of *D. melanogaster–D. simulans* $F_1$ hybrid females, we introgressed a new *D. simulans* fourth chromosome into an otherwise *D. melanogaster* background without use of radiation (Materials and Methods).

Hybrid male fertility in the new *4-sim* introgression line was scored by both sperm motility and number of offspring sired by individual males (Materials and Methods). Heterozygous *4-sim* males produce abundant motile sperm, whereas homozygous *4-sim* males typically produce immotile sperm (Table 1); sterile hybrid males thus show the same spermatogenic phenotype as those described previously (*5*). Although pure species *D. melanogaster* and heterozygous *4-sim* males ($ey^D$/*4-sim*) produce many offspring [232.1 ± 21.5 (SEM), $N = 13$, and 157.7 ± 13.9, $N = 15$, respectively], homozygous *4-sim* males produce none (0.0 ± 0.0, $N = 39$). In females, the *4-sim* chromosome has no effect on fertility. The *D. simulans* fourth chromosome thus causes true hybrid male sterility.

By using chromosomal deficiencies, we confirmed previous results (*5*, *7*) showing that the gene(s) causing hybrid male sterility resides within *Df(4)M101-62f*, which includes the proximal-most 21 genes on chromosome *4* (*10*). We dissected this region further by using deficiencies and genomic sequence data unavailable to earlier workers (Fig. 1). None of the new deficiencies uncovers hybrid sterility (Table 1 and Fig. 1). Assuming that *4-sim* hybrid sterility is caused by a single gene, we excluded all loci distal to *cubitus interruptus* as the cause of sterility. We also excluded *plexinB* on the basis of complementation tests. Our results thus show that one of the requisite loci for *4-sim* hybrid male sterility lies proximal to *plexinB* even if multiple genes within *Df(4)M101-62f* are involved (Fig. 1).

*JYAlpha* (*CG17923*), a 4.1-kb gene that encodes the alpha subunit of a $Na^+$ and $K^+$ adenosine triphosphatase ($Na^+/K^+$ ATPase), a transmembrane protein involved in ion exchange (*11*), was identified as a strong candidate from the four remaining loci in the region. Four mammalian isoforms of the $Na^+/K^+$ ATPase alpha subunit exist (*12*). One of these, α4, is expressed exclusively in testes (*13*) and is essential for sperm motility (*14*). JYAlpha from *D. melanogaster* ($JYAlpha^{mel}$) shows ~60% amino acid identity to mouse α4.

To test whether *JYAlpha* is the cause of *4-sim* hybrid male sterility, we performed complementation tests with the *4-sim* chromosome by using a *P*-element insertion in $JYAlpha^{mel}$ ($P\{y^+,w^+\}JYAlpha$). Because $P\{y^+,w^+\}JYAlpha$ does not appear to be a null mutation (Table 2), we remobilized the element. Two of the resulting excisions were chosen for analysis. $JYAlpha^{mel.8c}$ truncated $JYAlpha^{mel}$ after the first 268 amino acids, excluding the presumed active site. $JYAlpha^{mel.12a}$ restored wild-type sequence at $JYAlpha^{mel}$. $JYAlpha^{mel.8c}$/*4-sim* males were sterile, and their sperm motility resembled that of *Df(4)M101-62f/4-sim* (Table 2). As expected,

**Table 1.** Hybrid male sterility and deficiency mapping. $ey^D$ indicates *eyeless*-Dominant mutation.

| Genotype | Sperm motility | | | $\chi^2$ |
|---|---|---|---|---|
| | Many | Few | None | |
| $ey^D$/*4-sim* | 43 | 50 | 18 | 96.31*** |
| *4-sim*/*4-sim* | 0 | 32 | 92 | |
| *Df(4)M101-62f*/$ey^D$ | 110 | 175 | 36 | 289.2*** |
| *Df(4)M101-62f*/*4-sim* | 0 | 59 | 211 | |
| *Df(4)G*/$ey^D$ | 87 | 71 | 11 | 3.22 |
| *Df(4)G*/*4-sim* | 146 | 118 | 8 | |
| *Df(4)ED6369*/$ey^D$ | 29 | 8 | 0 | 1.10 |
| *Df(4)ED6369*/*4-sim* | 82 | 15 | 1 | |
| *Df(4)ED6366*/$ey^D$ | 105 | 36 | 1 | 2.47 |
| *Df(4)ED6366*/*4-sim* | 132 | 66 | 2 | |
| *Df(4)ED6364*/$ey^D$ | 205 | 34 | 1 | 9.09* |
| *Df(4)ED6364*/*4-sim* | 125 | 43 | 2 | |
| *Df(4)Δ3M*/$ey^D$ | 112 | 13 | 0 | 4.63 |
| *Df(4)Δ3M*/*4-sim* | 94 | 22 | 1 | |
| *Df(4)Δ9M*/$ey^D$ | 131 | 11 | 0 | 1.98 |
| *Df(4)Δ9M*/*4-sim* | 99 | 12 | 1 | |

***$P \ll 0.0001$. *Although marginally significant ($P < 0.05$), this deficiency does not uncover the severe "none" sperm motility phenotype seen in *4-sim* hybrids.

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*JYAlpha*$^{mel.12a}$/*4-sim* males were fully fertile (Table 2). *JYAlpha* is thus both necessary and sufficient for hybrid male sterility. Further analysis shows that *JYAlpha* is essential for sperm motility within *D. melanogaster* (Table 2).

Genes causing hybrid incompatibilities often evolve rapidly and show population genetic signs of divergence under positive natural selection (*3*, *15*–*17*). However, preliminary analysis of *JYAlpha* revealed an apparent difference in its location between *D. simulans* and *D. melanogaster*. A BLAST search of the *D. simulans* whole genome assembly (*18*) suggests that *JYAlpha* is flanked proximally by *CG9766* and distally by *complexin* (*cpx*), two loci that reside on the right arm of the third chromosome (*3R*) in *D. melanogaster*, *D. simulans*, and their sister species (Fig. 2A). This *3R*-linked locus represents the single best BLAST hit for *JYAlpha* (reciprocal e values = 0.0) and appears to be *JYAlpha*'s only location in *D. simulans*. We confirmed these results in several ways.

To determine *JYAlpha*'s chromosomal location in *D. simulans*, we performed crosses to track genetically the chromosome with which *JYAlpha* segregates. Strain-specific molecular markers in *JYAlpha*$^{sim}$ confirmed that the locus segregates with chromosome *3* (Materials and Methods).

To confirm *JYAlpha*'s precise location within the third chromosome of *D. simulans*, we attempted to amplify polymerase chain reaction (PCR) product across the putative *3R-4* breakpoints from pure *D. simulans C167.4*, control pure *D. melanogaster*, and homozygous *4-sim* hybrids. Amplification across both the proximal (*CG9766-JYAlpha*$^{sim}$) and the distal (*JYAlpha*$^{sim}$*-cpx*) breakpoints succeeded in pure *D. simulans* but failed in pure *D. melanogaster* and in homozygous *4-sim* hybrids, as expected if *JYAlpha* resides on *3R* in *D. simulans* but on *4* in *D. melanogaster* (Fig. 2B).

Next, we sequenced a large region of *3R* from pure *D. simulans.* In particular, we sequenced ~9.8 kb from *D. simulans C167.4*; this region extends from *CG9766* proximally to *cpx* distally. In *D. melanogaster*, *CG9766* and *cpx* are adjacent genes on *3R* (*19*). In *D. simulans*, however, the region between *CG9766* and *cpx* is interrupted by *JYAlpha* (Fig. 2A). *JYAlpha* is the only gene found on chromosome *4* of *D. melanogaster* that resides in this region of *3R* in *D. simulans* (fig. S2).

We also attempted to PCR-amplify *JYAlpha* from homozygous *4-sim* hybrids. We were unable to PCR-amplify *JYAlpha* product from homozygous *4-sim* hybrids with use of any primer pairs, despite routine amplification from pure-species *D. melanogaster* and *D. simulans* individuals. This confirms that an intact *JYAlpha* locus does not exist on the *D. simulans* fourth chromosome.

Lastly, we asked whether *JYAlpha* is single-copy. We already possess strong genomic and genetic evidence that *D. melanogaster* carries a single functional copy of *JYAlpha* (*10*) (Table 2). We also performed a Southern blot analysis of pure *D. melanogaster*, pure *D. simulans*, and homozygous *4-sim* flies. As expected from genome sequence data, *JYAlpha* appears to be single-copy in both species (fig. S3). Also as

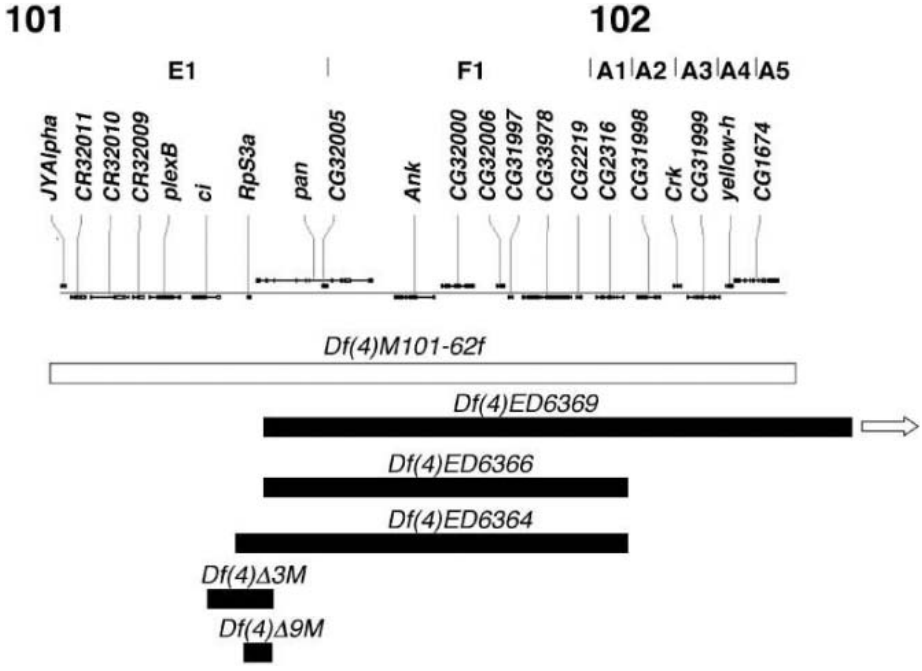


**Fig. 1.** Chromosome *4* region uncovered by *Df(4)M101-62f*. Horizontal bars represent deficiencies. White bars show deficiencies that uncover hybrid sterility when heterozygous with *4-sim*; black bars show deficiencies that fail to uncover sterility when heterozygous with *4-sim*. *Df(4)G* (not pictured) lies distal to *Df(4)M101-62f* and uncovers region 102E2 to 102F2. The distal breakpoint of *Df(4)ED6369* extends beyond *Df(4)M101-62f*. Map adapted from Entrez Genomes Build 4.3 (*28*).

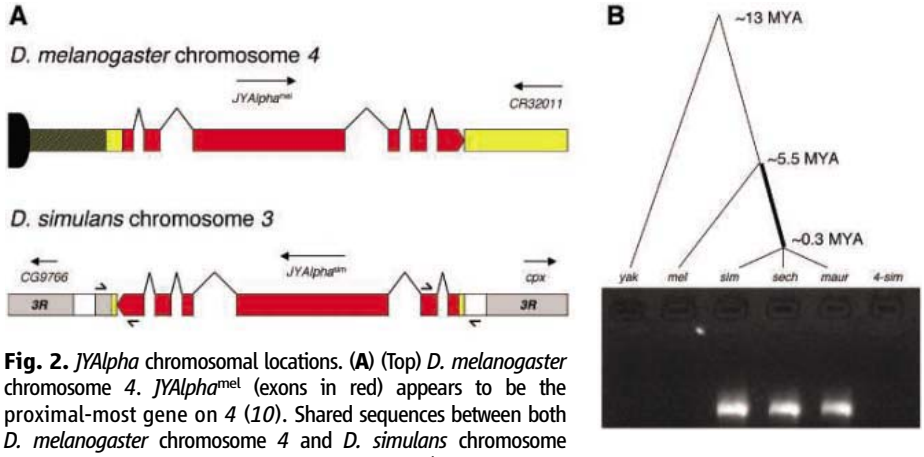


**Fig. 2.** *JYAlpha* chromosomal locations. (**A**) (Top) *D. melanogaster* chromosome *4*. *JYAlpha*$^{mel}$ (exons in red) appears to be the proximal-most gene on *4* (*10*). Shared sequences between both *D. melanogaster* chromosome *4* and *D. simulans* chromosome arm *3R* are coded yellow. Upstream of *JYAlpha*$^{mel}$, chromosome *4* becomes highly repetitive (striped) and then "centromeric" (black). Arrows indicate the direction of transcription. (Bottom) *D. simulans* chromosome arm *3R*. About 1.6 kb of sequence showing weak homology to sequence found on all major chromosomes (white) is present upstream and downstream of *JYAlpha*$^{sim}$ (red). This sequence shows no significant homology to known DNA-mediated transposable elements (*29*). *3R* material including *CG9766* and *cpx* is shown in gray. Half arrows give approximate primer locations for PCR across the *3R-4* breakpoints. Coding sequence between *JYAlpha*$^{mel}$ and *JYAlpha*$^{sim}$ shows no signature of divergence by positive selection, at least as crudely measured by the ratio of amino-acid changing to non-amino-acid changing substitutions: $K_a/K_s = 0.05$, consistent with purifying selection. (**B**) PCR-amplified region across the *3R-4 CG9766-JYAlpha*$^{sim}$ breakpoint. Analogous results were obtained from the *JYAlpha*$^{sim}$*-cpx* breakpoint. Divergence times are shown at speciation events on the phylogeny. The branch onto which the *JYAlpha* transposition event maps is bolded. *yak* indicates *D. yakuba*; *mel*, *D. melanogaster*; *sim*, *D. simulans*; *sech*, *D. sechellia*; *maur*, *D. mauritiana*; and *4-sim*, *4-sim* homozygotes.

expected, no hybridization was observed in our Southern blot for homozygous *4-sim* hybrids (fig. S3). This again demonstrates that no intact *JYAlpha* resides on chromosome *4* of *D. simulans*.

The cause of *4-sim* hybrid male sterility appears, therefore, to be surprisingly simple. A copy of *JYAlpha* exists on the fourth chromosome of *D. melanogaster* but not of *D. simulans*. Thus, a heterozygous *4-sim* hybrid male carries one *D. melanogaster* chromosome *4* and remains fertile, whereas a homozygous *4-sim* hybrid male lacks *JYAlpha* and is sterile. The sterility of this hybrid genotype reflects the complete absence of a locus essential for male fertility.

To determine the evolutionary direction of *JYAlpha*'s transposition, we performed PCR assays across the *3R-4* breakpoints in several species closely related to *D. melanogaster*. PCR amplification across both the proximal and distal break points succeeded in *D. sechellia* and *D. mauritiana*, showing that these species also carry *JYAlpha* on *3R* (Fig. 2B). The *D. sechellia* genome also confirmed that *JYAlpha* resides on *3R* in this species [Supporting Online Material (SOM) Text and fig. S2]. Amplification across the *3R-4* breakpoints did not succeed, however, in *D. yakuba*. Consistent with this, genome sequence data show that *JYAlpha* resides on the fourth chromosome in this species (SOM Text). Thus, *JYAlpha* appears to have resided ancestrally on chromosome *4* and was transposed to chromosome arm *3R* after the split of *D. melanogaster* from the *simulans* clade species but before the split of *D. simulans* from its sister species. This dates *JYAlpha*'s transposition to roughly 0.3 to 5.5 million years ago (*20*, *21*).

The transposition of *JYAlpha* raises several evolutionary questions. Because the transposition is evolutionarily old (at least $\sim 3 \times 10^6$ generations), it is unlikely that population genetic data would allow detection of a selective sweep associated with this event (*22*). Although the coding region of *JYAlpha* shows no obvious signs of divergence by positive natural selection between *D. melanogaster* and *D. simulans* (Fig. 2A legend), we cannot exclude a history of selection at this locus. Similarly, we cannot infer the exact mechanism of *JYAlpha*'s transposition. We can, however, rule out retroposition, because *JYAlpha*$^{sim}$ possesses introns (Fig. 2A).

It has been hypothesized that movement of gene function between chromosomes might cause postzygotic isolation, either by simple transposition or translocation (*1*) or by gene duplication-transposition followed by divergent evolution (*23*). Although gene duplication-transposition events are fairly common in *Drosophila* (*24*–*26*), our results suggest that *JYAlpha* is currently single-copy in both *D. melanogaster* and *D. simulans*. But it seems likely that a *JYAlpha* duplication existed sometime during the evolutionary history of the *simulans* clade. In any case, *JYAlpha* represents a clear example of a gene transposition causing reproductive isolation.

These findings raise the possibility that gene transposition could be important in the evolution of reproductive isolation. Although the present example sterilizes only a fraction of $F_2$ hybrids and has no effect on the fertility of $F_1$ hybrids, an analogous gene transposition event between the sex chromosomes could sterilize or kill $F_1$ hybrids between allopatric populations. If, for example, a *Y*-linked gene essential for male fertility transposed to the *X* chromosome, crosses between transposed and nontransposed populations would yield sterile $F_1$ hybrid males in one direction of the hybridization, consistent with Haldane's rule (*27*). Similarly, transpositions between sex chromosomes and autosomes, or between autosomes, could affect a fraction of hybrid backcross or $F_2$ genotypes (*23*). Gene transposition events between chromosomes need not, therefore, be common to have a large effect on hybrid fitness, because any hybrid that lacks a single essential gene would be inviable or sterile. The transposition of essential genes could represent a largely overlooked cause of reproductive isolation.

**Table 2.** *JYAlpha* complementation tests. *ci*$^D$, *cubitus interruptus*-Dominant mutation; *ey*$^D$, *eyeless*-Dominant mutation.

| Genotype | Sperm motility | | | $\chi^2$ |
|---|---|---|---|---|
| | Many | Few | None | |
| *ci*$^D$/*4-sim* | 91 | 45 | 10 | 173.1*** |
| *P*{*y*$^+$,*w*$^+$}*JYAlpha*/*4-sim* | 18 | 110 | 148 | |
| *P*{*y*$^+$,*w*$^+$}*JYAlpha*/*ey*$^D$ | 71 | 61 | 27 | 115.7*** |
| *P*{*y*$^+$,*w*$^+$}*JYAlpha*/*4-sim* | 6 | 114 | 116 | |
| *P*{*y*$^+$,*w*$^+$}*JYAlpha*/*ci*$^D$ | 186 | 20 | 2 | 313.1*** |
| *P*{*y*$^+$,*w*$^+$}*JYAlpha*/*P*{*y*$^+$,*w*$^+$}*JYAlpha* | 17 | 174 | 57 | |
| *ci*$^D$/*4-sim* | 94 | 25 | 3 | 173.4*** |
| *JYAlpha*$^{mel.8c}$/*4-sim* | 0 | 53 | 78 | |
| *JYAlpha*$^{mel.12a}$/*ci*$^D$ | 67 | 13 | 0 | 2.91 |
| JYAlpha$^{mel.12a}$/4-sim | 90 | 9 | 1 | |
| *JYAlpha*$^{mel.8c}$/*ci*$^D$ | 112 | 33 | 3 | 151.7*** |
| *JYAlpha*$^{mel.8c}$/*JYAlpha*$^{mel.8c}$ | 0 | 37 | 53 | |
| *JYAlpha*$^{mel.12a}$/*ci*$^D$ | 67 | 18 | 5 | 8.35* |
| *JYAlpha*$^{mel.12a}$/*JYAlpha*$^{mel.12a}$ | 92 | 8 | 2 | |

***$P \ll 0.0001$. *Although marginally significant ($P < 0.05$), the effect is in the wrong direction and probably reflects a mild *ci*$^D$ marker effect.


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**Supporting Online Material**

www.sciencemag.org/cgi/content/full/313/5792/1448/DC1
Materials and Methods
SOM Text
Figs. S1 to S3
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# Washing Away Your Sins: Threatened Morality and Physical Cleansing

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Physical cleansing has been a focal element in religious ceremonies for thousands of years. The prevalence of this practice suggests a psychological association between bodily purity and moral purity. In three studies, we explored what we call the "Macbeth effect"—that is, a threat to one's moral purity induces the need to cleanse oneself. This effect revealed itself through an increased mental accessibility of cleansing-related concepts, a greater desire for cleansing products, and a greater likelihood of taking antiseptic wipes. Furthermore, we showed that physical cleansing alleviates the upsetting consequences of unethical behavior and reduces threats to one's moral self-image. Daily hygiene routines such as washing hands, as simple and benign as they might seem, can deliver a powerful antidote to threatened morality, enabling people to truly wash away their sins.

When we find ourselves in morally compromising situations, how do we deal with the consequences of unethical behavior, given that most if not all of us desire a moral self-image? This paper investigates a basic coping mechanism that has been used by religions for centuries: washing away one's sins.

Physical cleansing, such as bathing or washing hands, is at the core of many religious rituals. Baptism, for instance, is a water purification ritual practiced by Christians, Mandaeanists, and Sikhs. Christians follow the admonition, "Arise and be baptized, and wash away your sins" (*1*), with faith that through the symbolic cleansing of their bodies they might also achieve a cleansing of conscience. Physical cleansing is also central to Islam; wudu (often translated as "ablution") is the Muslim act of washing parts of the body in clean water to prepare for worship. Likewise, Hinduism requires great attention to bodily purity (*2*). Thus, many major religions discipline bodily purity, suggesting that physical cleansing ceremonies can purify the soul.

Research on the correspondence between physical and moral purity (*3*) has speculated that people are predisposed to use categories that are based on bodily experience (such as clean versus dirty) to construct complex social categories (such as moral versus immoral) (*4*). For example, in English, words such as "clean" and "pure" describe both physical and moral states (e.g., he has a clean record). Likewise, the Mandarin phrase "a pair of dirty hands" refers to a person who steals.

The association between bodily and moral purity may be based not only in cognition, but in emotion as well. As an example, "disgust" represents an emotion that is experienced in both physical and moral domains. Pure disgust was originally a gustatory emotion rooted in evolution to avoid the intake of potentially hazardous food. Over time, it has taken on social and cultural meanings and has expanded to encompass broader categories of aversions including social or moral violations (*5*, *6*). Although the experience of pure disgust devoid of moral connotations can be subjectively and behaviorally differentiated from the experience of disgust with moral connotations (*7*), they coincide considerably. Specifically, previous research suggests that pure disgust and moral disgust not only lead to similar facial expressions and physiological activation (*6*) but also recruit partially overlapping brain regions, mainly in the frontal and temporal lobes (*7*). Given the psychological, physiological, and neurological overlap between physical and moral disgust, physical cleansing acts that mitigate physical disgust might also reduce social or moral disgust, thereby alleviating moral condemnation.

Thus, Lady Macbeth's hope that a little bit of water would clear her of the treacherous murder of King Duncan might not have been a product of literary creativity, but of Shakespeare's acute understanding of the human psyche. If physical and moral purity are so psychologically intertwined, Lady Macbeth's desperate obsession with trying to wash away her bloodied conscience while crying, "Out, damned spot! Out, I say!" (*8*) may not have been entirely in vain.

Given that physical cleansing might function as a surrogate for moral purification, we set out to investigate (i) whether a threat to moral purity activates a need for physical cleansing (i.e., the Macbeth effect) and (ii) whether physical cleansing is actually efficacious in helping people cope with moral threats. We first determined whether a threat to moral purity increases the mental accessibility of cleansing-related words. We asked participants to recall in detail either an ethical or unethical deed from their past and to describe any feelings or emotions they experienced. Then they engaged in a word completion task in which they converted word fragments into meaningful words (*9*). Of the six word fragments, three (W _ _ H, SH _ _ ER, and S _ _ P) could be completed as cleansing-related words (wash, shower, and soap) or as unrelated words (e.g., wish, shaker, and step). Participants who recalled an unethical deed generated more cleansing-related words than those who recalled an ethical deed [$F(1,58) = 4.26$, $P = 0.04$], suggesting that unethical behavior enhances the accessibility of cleansing-related concepts (Table 1).

Was this accessibility the result of an urge to cleanse one's body when moral integrity was threatened? Study 2 investigated whether an implicit threat to moral purity produces a psychological desire for cleansing, through expressed preferences for cleansing products. Participants were told that we were investigating the relationship between handwriting and personality and were asked to hand-copy a short story written in the first person. The story described either an ethical, selfless deed (helping a co-worker) or an unethical act (sabotaging a co-worker) (*9*). Participants then rated the desirability of various products from 1 (completely undesirable) to 7 (com-

**Table 1.** Summary of Results. Study 1 measured the effect of recalling ethical versus unethical behavior on the mental accessibility of cleansing-related words. Study 3 explored the effect of recalling ethical versus unethical behavior on the likelihood of choosing antiseptic wipes (over pencils). Study 4 assessed the effect of hand cleansing on the likelihood of engaging in moral compensatory behaviors (i.e., offering help).

| Study 1: Average number of cleansing-related words completed (SEM) | | Study 3: Percentage who chose antiseptic wipes | | Study 4: Percentage who volunteered to help | |
|---|---|---|---|---|---|
| Ethical recall ($n = 30$) | Unethical recall ($n = 30$) | Ethical recall ($n = 16$) | Unethical recall ($n = 16$) | Cleansed ($n = 22$) | Not cleansed ($n = 23$) |
| .90 (1.88) | 1.43 (1.77) | 33.3% | 66.7% | 40.9% | 73.9% |

[1]Department of Organizational Behavior and HR Management, Joseph L. Rotman School of Management, University of Toronto, Toronto, Ontario M5S 3E6, Canada. [2]Department of Management and Organizations, Kellogg Graduate School of Management, Northwestern University, Chicago, IL 60208, USA.

*To whom correspondence should be addressed. E-mail: chenbo.zhong@rotman.utoronto.ca

pletely desirable). Cleansing products included Dove shower soap, Crest toothpaste, Windex cleaner, Lysol disinfectant, and Tide detergent; other products included Post-it Notes, Nantucket Nectars juice, Energizer batteries, Sony CD cases, and Snickers bars. As expected, copying the unethical story increased the desirability of cleansing products as compared to copying the ethical story [$F(1,25) = 6.99$, $P = 0.01$], with no differences between conditions for the noncleansing products [$F(1,25) = 0.02$, $P = 0.89$] (Fig. 1).

We sought to replicate the results of Study 2 using behavioral measures, so our next study examined the likelihood of taking an antiseptic cleansing wipe after recalling an ethical or unethical deed. Participants engaged in the same recall task as in Study 1 and were then offered a free gift and given a choice between an antiseptic wipe and a pencil (verified in a control condition to be equally attractive offerings). Those who recalled an unethical deed were more likely to take the antiseptic wipe (67%) than were those who recalled an ethical deed (33%) ($\chi^2 = 4.57$, $P = 0.03$) (Table 1).

These three studies provided evidence for the Macbeth effect: Exposure to one's own and even to others' moral indiscretions poses a moral threat and stimulates a need for physical cleansing. Our final study investigated the efficacy of physical cleansing—can it actually wash away moral sins?

Physical cleansing may wash away moral sins through symbolic self-completion (*10*); that is, people are motivated to complete their self-definitions (e.g., musicians) when indicators or symbols of this definition are lacking (e.g., skills) by engaging in activities that complete the symbols (e.g., training). Thus, when moral self-definition is at stake, such as when one has indulged in morally questionable activities, one should naturally be motivated to engage in activities that will restore moral integrity. For instance, Tetlock and colleagues (*11*) have shown that the mere contemplation of violating one's core values spurs intent to take actions that will restore and protect those values. The restoration or completion of the moral self can be achieved through direct restitution, but it may also be achieved through substitutable symbols or activities that are not directly related (*10*, *11*). Given the demonstrated association between physical cleansing and moral purity, cleansing activities that improve physical cleanliness may also compensate for moral impurity.

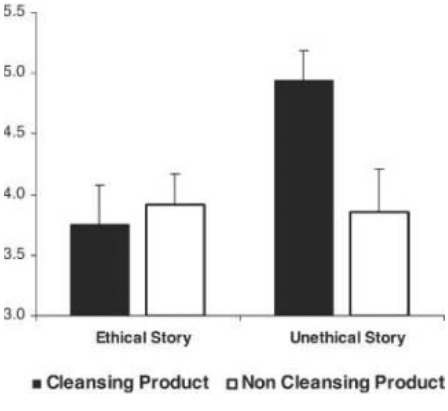


**Fig. 1.** Effect of hand-copying an ethical ($n = 16$) vs. unethical story ($n = 11$) on the desirability of cleansing and noncleansing products on a scale of 1 (low) to 7 (high). Error bars represent standard error.

Thus, we expected that a threat to the moral self would motivate the restoration of moral purity through direct compensatory behaviors (e.g., volunteering to help). If, however, physical cleansing restores the moral self, then individuals should have less need to engage in direct compensatory behaviors after physically cleansing themselves.

This is indeed what we found. In Study 4, participants described an unethical deed from their past (the same recall task as in Study 1). Afterwards, they either cleansed their hands with an antiseptic wipe or not. Then they completed a survey regarding their current emotional state (*9*). After completing the survey, participants were asked if they would volunteer without pay for another research study to help out a desperate graduate student. Presumably, participants who had cleansed their hands before being solicited for help would be less motivated to volunteer because the sanitation wipes had already washed away their moral stains and restored a suitable moral self.

As predicted, physical cleansing significantly reduced volunteerism: 74% of those in the not-cleansed condition offered help, whereas only 41% of participants who had a chance to cleanse their hands offered help ($\chi^2 = 5.02$, $P = 0.025$). Thus, the direct compensatory behavior (i.e., volunteering) dropped by almost 50% when participants had a chance to physically cleanse after recalling an unethical behavior (Table 1).

Physical cleansing also influenced participants' emotional state. Based on an exploratory factor analysis (*9*), the assessed emotions clustered into two categories: moral emotions (i.e., disgust, regret, guilt, shame, embarrassment, and anger; Cronbach Alpha = 0.90) and nonmoral emotions (i.e., confidence, calm, excitement, and distress; Cronbach Alpha = 0.65). As expected, participants who cleansed their hands after the unethical recall reported reduced moral emotions ($M = 1.75$, SEM = 0.19) compared with those who did not ($M = 2.23$, SEM = 0.26), $F(1,41) = 2.94$, $P = 0.047$. Hand washing, however, did not influence nonmoral emotions, $F(1,41) = 0.25$, $P = 0.31$ (*12*).

These four studies document a psychological association between physical and ethical cleanliness: Threats to moral purity activate a need for physical cleansing, which can assuage moral emotions and reduce direct compensatory behaviors. Although there are surely limits to the absolution afforded by a bar of soap, our findings shed light on Lady Macbeth's feverish attempts to physically cleanse herself after the murder of King Duncan. If even an implicit threat to one's moral image can produce a psychological need to engage in cleansing behaviors, it is only natural that those who suffer genuine guilt would be all the more relentless in their attempts to restore a pure conscience.

The implications of this research may be substantial. Future studies that specifically address the psychological and behavioral consequences of physical cleanliness will provide valuable insight into regulatory mechanisms that drive ethical decisions. Given the boost to one's moral self afforded by physical cleansing, how might it influence subsequent behavior? Would adherence to a rigorous hygiene regimen facilitate ethical behavior? Or, would cleansing ironically license unethical behavior? It remains to be seen whether clean hands really do make a pure heart, but our studies indicate that they at least provide a clean conscience after moral trespasses.

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12. We included participants' sex as a covariate. Sex itself had no impact on emotional state or offers of help. We used one-tailed tests for the effect of cleansing on emotional state because we had predicted that hand washing would reduce moral emotions but not affect the other emotions.
13. This research was supported by a grant from Dispute Resolution Research Center at the Kellogg School of Management at Northwestern University and an NSF Graduate Fellowship to K.L. We thank A. Galinsky for his insights and support and G. Ku and K. Murnighan for their thoughtful comments on earlier drafts of the paper.

### Supporting Online Material

www.sciencemag.org/cgi/content/full/313/5792/1451/DC1
Materials and Methods
Tables S1 and S2
References and Notes

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10.1126/science.1130726

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## POSITIONS OPEN

### FACULTY POSITION IN CANCER CELL BIOLOGY
**Department of Biological Sciences**
**Purdue University**

The Department of Biological Sciences invites applications for a tenure-track faculty position in cancer cell biology. We are seeking candidates who use mammalian model systems to address fundamental questions related to cancer stem cell biology and tumor development. We expect to fill an academic year appointment at the **ASSISTANT PROFESSOR** level; however appointment at a higher rank will be considered for qualified applicants.

The Department of Biological Sciences **(website: http://www.bio.purdue.edu/)** has over 50 faculty members directing research in a wide range of fields including bioinformatics, molecular, cellular, and developmental biology, and ecology. Eighteen faculty are members of the NCI-designated Purdue Cancer Center **(website: http://www.cancer.purdue.edu/)** and of the interdisciplinary Oncological Sciences Center, a key component of Purdue's Discovery Park **(website: http://www.purdue.edu/discoverypark/).** The Department of Biological Sciences operates a state-of-the-art Transgenic Mouse Core Facility in conjunction with the Purdue Cancer Center. Over the next several years, we anticipate additional faculty positions in integrative disease biology, comparative oncology, developmental biology, and molecular evolution as the University expands the Life Sciences on campus. Several new buildings, including the Biomedical Engineering Building and the Bindley Bioscience Center, house shared facilities for cell sorting, image analysis, genomics, quantitative and functional proteomics, and other biological instrumentation. A new Structural Biology Building will consolidate Purdue's world-class expertise in X-ray crystallography, cryo-electron microscopy and nuclear magnetic resonance.

The successful cancer cell biology applicant must have a Ph.D. or equivalent in an appropriate discipline and at least two years of postdoctoral experience. Applicants should demonstrate a strong potential for excellence in research, the promise of extramural funding, and a commitment to excellence in teaching. Applications must be submitted electronically as a PDF file that includes detailed curriculum vitae, the names and addresses of three referees, a summary of research interests, and a one-paragraph teaching statement to **e-mail: chairccb@bio.purdue.edu.** Inquiries should be directed to: **Professor Elizabeth J. Taparowsky, Chair, Cancer Cell Biology Search Committee, Department of Biological Sciences, Purdue University, 915 W. State Street, West Lafayette, IN 47907-2054.** Review of applications will begin on October 1, 2006, and will continue until a suitable pool of applicants has been identified.

The Department also plans to fill, in a college-wide effort called COALESCE (Cooperative Areas Linking and Extending Science), positions in multidisciplinary areas, including membrane science, bioinformatics, and nanoscience. Information and application criteria for these positions may be obtained at **website: http://www/science.purdue.edu/COALESCE/.** Applicants to one search may be included in other relevant searches when appropriate.

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### UNIVERSITY OF CALIFORNIA, MERCED
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## POSITIONS OPEN

### FACULTY POSITION in VERTEBRATE DEVELOPMENTAL BIOLOGY
**Department of Biological Sciences**
**Purdue University**

The Department of Biological Sciences invites applications for a tenure-track faculty position in Vertebrate Developmental Biology. Primary consideration will be given to candidates who use zebrafish as a genetic model system to ask fundamental questions in developmental biology. We expect to fill an academic-year appointment at the **ASSISTANT PROFESSOR** level; however appointment at a higher rank will be considered for qualified applicants.

The Department has over 50 faculty members directing research in a wide range of fields from bioinformatics, through molecular and systems biology, to evolutionary biology and ecology. Over the next several years we anticipate adding faculty positions in developmental biology, integrative disease biology, and molecular evolution. The Department plans to build a common zebrafish facility that will service five laboratories. Further information about the Department is available at **website: http://www.bio.purdue.edu/.** Purdue University is expanding its Life Sciences Program and as part of this initiative there are several new buildings completed or planned. These include Biomedical Engineering, Structural Biology, and the Bindley Bioscience Center which houses shared facilities for image analysis, genomics, quantitative and functional proteomics, and other biological instrumentation.

The successful vertebrate developmental biology applicant must have a Ph.D. or equivalent in an appropriate discipline and at least two years of postdoctoral experience. We seek applicants with a strong potential for excellence in research, the ability to attract extramural funding, and a commitment to excellence in teaching. Applications must be submitted electronically as a PDF file that includes detailed curriculum vitae, the names and addresses of three referees, a summary of research interests, and a one to two-paragraph description of teaching philosophy and interests to **e-mail: chair_devo@bio.purdue.edu.** Please arrange to have three letters of recommendation sent to the same e-mail address. Inquiries should be directed to: **Professor Donna M. Fekete, Chair of Developmental Search Committee, Department of Biological Sciences, Purdue University, 915 W. State Street, West Lafayette, IN 47907-2054.** Review of completed applications will begin September 30, 2006, and will continue until a suitable pool of applicants has been identified.

The Department also plans to fill, in a college-wide effort called COALESCE (Cooperative Areas Linking and Extending Science), a number of other biology faculty positions in multidisciplinary areas, including membrane science, bioinformatics, and nanoscience. Applicants in these fields may apply directly to **website: http://www.science.purdue.edu/COALESCE/.** Applicants to one search may be included in other relevant searches when appropriate. *Purdue University is an Equal Opportunity/Equal Access/Affirmative Action Employer and is committed to building a diverse faculty of excellence.*

### ASSISTANT PROFESSORSHIP IN CHEMISTRY
**Harvard University**
**Department of Chemistry and Chemical Biology**

Applicants are invited to apply for tenure-track Assistant professorships in all fields of chemistry. Applicants should arrange to have three letters of recommendation sent independently and should provide curriculum vitae, a list of publications, and an outline of their future research plans. Applications and supporting materials should be sent to: **Chair, c/o Ms. Carol Gonzaga, Department of Chemistry and Chemical Biology, Harvard University, 12 Oxford Street, Cambridge, MA 02138-2902.** Reference position: JFCCB133B. The deadline date for receipt of applications and supporting materials is October 15, 2006. *Harvard University is an Affirmative Action, Equal Opportunity Employer. Applications from and nominations of women and minority candidates are strongly encouraged.*

## POSITIONS OPEN

### IMMUNOLOGIST

Simon Fraser University is developing an integrated graduate program in infectious diseases (ID), to be mounted jointly by the Faculty of Science (FS) and the new Faculty of Health Sciences. A tenure-track faculty position is available in the Department of Molecular Biology and Biochemistry, FS, at the **ASSISTANT PROFESSOR** level; under exceptional circumstances appointment might be at a higher rank. Research expertise is preferred in protective humoral immunity against infectious disease. Laboratory space, including a BCL-2/P2 bio-containment facility, will be available. Applicants should have postdoctoral research experience; teaching experience is a plus. The successful applicant will be expected to establish a funded research program, teach at the undergraduate and graduate levels, and contribute to development of the ID program.

Applicants should submit curriculum vitae, three letters of reference, up to four representative publications, and statements of research interests/plans and teaching philosophy to:

**Immunology Search Committee**
**Molecular Biology and Biochemistry**
**Simon Fraser University**
**Burnaby, British Columbia, V5A 1S6 Canada**
**E-mail: mbbchair@sfu.ca.**

Review of applications will begin on November 1, 2006, and will continue until the position is filled. See the employment section of **website: http://www.sfu.ca/mbb/** for more information.

All qualified candidates are encouraged to apply; however, *Canadians and permanent residents will be given priority. Simon Fraser University is committed to an equity employment program that includes special measures to achieve diversity among its faculty and staff. We therefore particularly encourage applications from qualified women, First Nations people, persons with disabilities, and members of visible minorities.Under the authority of the University Act personal information that is required by the University for academic appointment competitions will be collected. For details see* ***website: http://www.sfu.ca/vpacademic/Faculty_Openings/Collection_Notice.html.***

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### ASSISTANT and ASSOCIATE/ FULL PROFESSOR
### Chemical Engineering

The Department of Chemical Engineering at the University of Delaware invites applications for two tenure-track faculty positions. We anticipate making one appointment at the Assistant Professor level and one at the Professor or Associate Professor level. The Department (**website: http://www.che.udel.edu**) is consistently ranked among the ten leading chemical engineering departments nationwide and has especially strong research programs in bioengineering, control, kinetics and catalysis, colloid and surface science, polymers and composites, thermodynamics, separations, and materials science. Requirements: Ph.D. or equivalent in chemical engineering or a related field. Applicants for the senior position are expected to have a commensurate record of accomplishments. Duties: Lead vigorous research programs, teach, and advise students at both the undergraduate and graduate levels. Applications in all research areas will be considered, with emphasis on candidates with expertise in biochemical and biological engineering.

Contact: Please send curriculum vitae, a description of research and teaching interests, and the names, addresses, telephone numbers, and e-mail addresses of three references in PDF format to: **Professor Norman Wagner, Search Committee Chairperson,** at **e-mail: che-faculty-search@udel.edu;** or mail to: **Professor Norman Wagner, Search Committee Chairperson, Department of Chemical Engineering, University of Delaware, Newark, DE 19716-3110.** PDF submissions are strongly encouraged. The committee will commence review of applications on November 15, 2006. The curriculum vitae and letters of reference shall be shared with Departmental faculty.

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## POSITIONS OPEN

WISCONSIN

### ASSISTANT PROFESSOR
### Translational Neuroscience
### University of Wisconsin, Madison

The University of Wisconsin (UW), Madison, recently made a significant commitment to develop a program in translational research for neurological diseases through its innovative Cluster Hiring Initiative **(website: http://www.waisman.wisc.edu/tr),** which provides permanent, full salary support for faculty positions in emerging fields of scholarship that do not fall entirely within traditional departmental disciplines and structures.

The current recruitment seeks to identify a tenure-track Assistant Professor interested in developing novel therapies for treating diseases of the brain, with a particular emphasis on neurodevelopmental disorders. Areas of interest might include, but are not limited to, gene discovery, neurochemical pathology, manipulation of the blood-brain barrier, gene therapy, stem cells, or animal models. The incumbent will join a thriving community of basic and clinical Neuroscientists supported by a campuswide Neuroscience Training Program and a Center for Neuroscience. Laboratory space will be located in newly remodeled facilities at the Waisman Center, which also includes rodent housing and a clinical-grade biotherapeutics manufacturing facility. Potential departmental affiliations include basic and clinical sciences depending on the candidate.

To ensure full consideration, please submit application materials by October 16, 2006. Applications will be accepted until the position is filled. Submit application materials electronically. Send a cover letter referring to position vacancy listing 53389, curriculum vitae, description of research goals, three publications, and three letters of reference to: **Albee Messing, V.M.D., Ph.D., Chair, Translational Neuroscience Cluster Search Committee, Waisman Center, e-mail: messing@waisman.wisc.edu.**

*UW Madison is an Equal Opportunity/Affirmative Action Employer. We promote excellence through diversity and encourage all qualified individuals to apply.*

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### FACULTY POSITION
### Pharmaceutical Biotechnology/Pharmaceutics

The Department of Pharmaceutical Sciences at North Dakota State University invites applications for a tenure-track position at the rank of **ASSISTANT/ ASSOCIATE PROFESSOR.** The appointment is expected to begin on or after August 15, 2007. Applicants must possess a Ph.D. degree in biotechnology, pharmaceutics, biomedical or biochemical engineering, or in a related field; have at least two years of postdoctoral experience with a strong record of scholarship. Preferred qualifications include expertise in pharmaceutical biotechnology or novel drug delivery of biotechnologically derived molecules and a degree in pharmacy. Successful candidates will be expected to teach pharmaceutical biotechnology and basic pharmaceutics in the Doctor of Pharmacy program, mentor M.S./Ph.D. students, and develop an extramurally funded research program.

A highly competitive salary and a startup package commensurate with qualifications and experience are available. Additional information concerning the Department, the University, and Fargo can be obtained at **website: http://www.ndsu.edu/pharmsci/.** Review of applications will begin on October 31, 2006, and will continue until the position is filled. The application portfolio containing the curriculum vitae, statement of teaching philosophy, description of research interests and future plans, and the names and contact information for three references should be submitted to: **Dr. Sanku Mallik, North Dakota State University, College of Pharmacy, Fargo, ND 58105, telephone: 701-231-7888, fax: 701-231-8333, e-mail: sanku.mallik@ndsu.edu.**

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## POSITIONS OPEN

### ASSISTANT PROFESSOR
### Evolutionary Biology, Santa Clara University

The Biology Department of Santa Clara University (SCU) is seeking applicants for a tenure-track Assistant Professor position. We seek a scientist with research and teaching interests in evolutionary biology. The candidate should be utilizing molecular-based approaches in their research. The SCU Biology Department offers well-equipped, modern facilities for classroom and laboratory teaching and research. Teaching responsibilities for this position may include a majors course in evolutionary biology with laboratory and/or field components, participation in the evolution/ecology/biodiversity segment of our introductory series for biology majors, and a nonmajors evolution course. People with research or teaching interests relevant to plant systems are especially encouraged to apply. This position requires a Ph.D., postdoctoral experience, and a strong commitment to mentoring undergraduates of diverse backgrounds. Applicants should provide current curriculum vitae, and a cover letter or separate statements describing: (1) research interests that would be pursued at SCU, (2) teaching philosophy and courses of interest, and (3) contributions the candidate would make to Santa Clara as a diverse and inclusive institution. Applications should be sent to: **Evolutionary Biology Search Committee, Biology Department, Santa Clara University, 500 El Camino Real, Santa Clara, CA 95053.** The applicant should also request at least two letters of reference be sent to this address as well. Applications, inquiries, and reference letters can also be sent by **e-mail: biology@scu.edu.** Applications must be received by November 3, 2006. Santa Clara University is the Catholic, Jesuit university of Silicon Valley, with a 155-year tradition of educational excellence. For more information about the Biology Department and Santa Clara University, see **website: http://www.scu.edu/biology.** *Santa Clara University is an Equal Opportunity/ Affirmative Action Employer, committed to excellence through diversity, and, in this spirit, particularly welcomes applications from women, persons of color, and members of historically underrepresented groups. The University will provide reasonable accommodations to all qualified individuals with a disability.*

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### ASSISTANT PROFESSOR, MARINE BIOLOGY
### Pacific Lutheran University

The Department of Biology at Pacific Lutheran University (PLU) invites applications for a tenure-track position in marine biology to begin 1 September 2007. Ph.D. required. Teaching responsibilities will include participation in introductory biology course(s) for majors/nonmajors; a broad-based marine ecology, biological oceanography, or marine biology course with a field-based laboratory; and, one or more additional courses related to the candidate's area of specialization. Research involving undergraduates is expected and supported. The Department of Biology has 14 full-time faculty members. Please submit your curriculum vitae, copies of undergraduate and graduate transcripts, a statement of teaching philosophy, and a summary of current research interests; also arrange to have three letters of recommendation sent on your behalf. Send all materials to: **Marine Biology Search Committee, c/o Dr. Dana Garrigan, Chair, Department of Biology, Pacific Lutheran University, Tacoma, WA 98447.** Review of applications will begin on 16 October 2006. Tacoma is located on Puget Sound in Washington, a uniquely scenic and biologically diverse region. Seattle and Mt. Rainier are about 40 miles from our suburban campus. Pacific Lutheran University enrolls 3,700 students and is committed to finding connections between the liberal arts and professional schools, and to promoting international education and undergraduate research. PLU enjoys a healthy and progressive relationship with the Evangelical Lutheran Church of America and values its tradition of academic freedom. The majority of our faculty has lived abroad or speaks a second language. For more information, please visit **website: http://www.nsci.plu.edu/biol/jobs.htm.** *In addition to having a diverse faculty, PLU serves a diverse clientele and is an Affirmative Action/Equal Opportunity Employer.*

## POSITIONS OPEN

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**Stanley S. Scott Cancer Center**
**Louisiana State University Health Sciences Center**
**New Orleans**

The Louisiana State University Health Sciences Center (LSUHSC), School of Medicine in New Orleans, Stanley S. Scott Cancer Center, is accepting applications for the Director of the Cancer Center. The position requires an M.D., Ph.D., or both, with combined M.D., Ph.D. degrees preferred. Preference will be given to candidates with experience in working within or in developing a successful plan for an NCI designated cancer center. The ideal incumbent must demonstrate scholarly experience as evidenced by academic accomplishments, publications, service on national study sections, service on editorial boards, and a track record of extramural research funded through NCI or other NIH funding mechanisms. Administrative experience with building either cancer research programs or cores in an academic setting is preferred. The incumbent should qualify for appointment at the level of Professor. A track record in basic, translational, epidemiological, and/or clinical research is required. Knowledge of current clinical oncology operations in relation to academic medicine will be viewed positively. The ideal candidate should have demonstrated ability in developing translational research from the basic scientific observations to clinical trials or clinically relevant research.

The incumbent will be appointed to the appropriate clinical department and will be a member of the Stanley S. Scott Cancer Center. Also the incumbent will be Co-Director of the Louisiana Cancer Research Consortium of LSU and Tulane. Please send curriculum vitae including current grant funding, a brief cover application letter detailing professional interests and goals, and the names of three references to: **Bernard Wan, Dean's Office, School of Medicine, 533 Bolivar Street, New Orleans, LA 70112** with an **e-mail: bwan@lsuhsc.edu.**

*LSUHSC is an Affirmative Action/Equal Opportunity Employer.*

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### ASSISTANT PROFESSOR, EVOLUTIONARY BIOLOGY
**Pacific Lutheran University**

The Department of Biology at Pacific Lutheran University (PLU) invites applications for a tenure-track position in evolutionary biology to begin 1 September 2007. Ph.D. required. Candidates with expertise in population, quantitative, or evolutionary genetics; molecular evolution; or molecular systematics are especially encouraged to apply. Teaching responsibilities will include participation in introductory biology course(s) for majors/nonmajors and one or more upper-division majors' courses related to the candidate's area of specialization. Research involving undergraduates is expected and supported. The Department of Biology has 14 full-time faculty members. Please submit your curriculum vitae, copies of undergraduate and graduate transcripts, a statement of teaching philosophy, and a summary of current research interests; also arrange to have three letters of recommendation sent on your behalf. Send all materials to: **Evolutionary Biology Search Committee, c/o Dr. Dana Garrigan, Chair, Department of Biology, Pacific Lutheran University, Tacoma, WA 98447.** Review of applications will begin on 31 October 2006. Tacoma is located on Puget Sound in Washington, a uniquely scenic and biologically diverse region. Seattle and Mt. Rainier are about 40 miles from our suburban campus. Pacific Lutheran University enrolls 3,700 students and is committed to finding connections between the liberal arts and professional schools and to promoting international education and undergraduate research. PLU enjoys a healthy and progressive relationship with the Evangelical Lutheran Church of America and values its tradition of academic freedom. The majority of our faculty has lived abroad or speaks a second language. For more information, please visit **website: http://www.nsci.plu.edu/biol/jobs.htm.** *In addition to having a diverse faculty, PLU serves a diverse clientele and is an Affirmative Action/Equal Opportunity Employer.*

## POSITIONS OPEN

### NEUROSCIENCE OPEN-RANK FACULTY POSITIONS
**Northwestern Feinberg School of Medicine**

The Davee Department of Neurology and the Northwestern University Institute for Neuroscience announce a new search to recruit outstanding individuals for full-time, tenure-track, appointments at the level of **ASSISTANT, ASSOCIATE,** or **FULL PROFESSOR.** Applications will be considered in areas of molecular/cellular neurobiology, especially development, biochemistry, stem cells, genetics, and neurodegeneration.

Applicants must include the following materials: (1) current curriculum vitae and list of publications, (2) brief statement of research interests (three pages or less), and (3) three letters of reference sent on their behalf to:

**Chair of the Neurology Search Committee**
**c/o Tara Y. Davis**
**Northwestern University**
**E-mail: tdavis@northwestern.edu**
**Websites: http://www.feinberg.northwestern.edu, and www.northwestern.edu/nuin**

Please refer to academic search numbers P-121-05 or P-122-05. Completed applications must be received by December 1, 2006. Appointments will commence on or after June 1, 2007.

*Northwestern University is an Equal Opportunity/Affirmative Action Educator and Employer and invites applications from all qualified individuals. Applications from women and minorities are especially sought.*

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### TENURE-TRACK POSITIONS IN BIOCHEMISTRY, ANALYTICAL CHEMISTRY
**Department of Chemistry and Biochemistry**
**California State University, Fullerton**

California State University, Fullerton, Department of Chemistry and Biochemistry seeks applicants for tenure-track faculty positions in biochemistry and in analytical chemistry, preferably at the **ASSISTANT PROFESSOR** level, to begin August 2007. Postdoctoral or equivalent research experience is preferred; women and underrepresented minority applicants are particularly welcome. A willingness to engage in collaborative research in related fields is desirable.

Successful applicants must have a Ph.D. in biochemistry or chemistry and have the potential to develop vigorous research programs involving undergraduate and graduate (M.S.) students that attract external funding and lead to refereed publications. Successful candidates must be committed to excellence in teaching a diverse population of students. Primary teaching responsibilities will be in the core lecture and laboratory courses of the discipline at the undergraduate level, and in graduate courses.

Information about the University and Department, as well as full information on the advertised positions is available online at **website: http://chemsrvr2.fullerton.edu/.**

Applicants should send a detailed curriculum vitae (including names of references), a summary of proposed research, and a summary of teaching philosophy and preferences, and arrange for three letters of recommendation from individuals familiar with their teaching and research potential. Please send these to:

**Dr. Maria C. Linder**
**Chair, Department of Chemistry and Biochemistry**
**California State University, Fullerton**
**P.O. Box 6866**
**Fullerton, CA 92834-6866**
**Telephone: 714-278-3621**
**Fax: 714-278-5316**
**E-mail: mlinder@fullerton.edu**

Review of applications will begin on October 1, 2006, and continue until the positions are filled.

*Cal State Fullerton is an Affirmative Action/Equal Opportunity/Title IX/ADA Employer.*

## POSITIONS OPEN

### TENURE-TRACK POSITION
**Plant Biology**
**Grinnell College, Department of Biology**

Grinnell College invites applications for a tenure-track faculty position in plant biology at the rank of **ASSISTANT PROFESSOR.** Candidates must have a Ph.D., postdoctoral experience, and plan an active research program involving undergraduates. The position will begin in August 2007. The successful candidate will be expected to teach three courses (with laboratories) in our inquiry-based curriculum: Introduction to Bilogical Inquiry (Biology 150); Molecules, Cells, and Organisms (Biology 251); and a third course at the upper level in the candidate's area of specialty. We seek candidates with a background in plant molecular biology, biochemistry, development, or physiology. The successful candidate should demonstrate an interest in participation in the College's general education offerings. Startup funds, excellent equipment, new and recently renovated facilities, support for student-faculty research, and a biological field station are available. Grinnell is a highly selective, residential, liberal arts college with an enrollment of approximately 1,500 students from across the country and around the world. One-third of the college's students major in the sciences, including about 30 biology and 15 biological chemistry graduates per year in recent years. The Department has 12 faculty with active research programs and offers an innovative curriculum centered around research-based learning; for information see **website: http://www.grinnell.edu/academic/biology/.** In their letters of application, candidates should address their ability to teach courses described above, and discuss their interest in developing as a teacher and scholar in an undergraduate, liberal arts environment that emphasizes close student-faculty interaction. Candidates should also address how they can contribute to diversity, a core value of the Grinnell College community. Please send curriculum vitae, copies of all transcripts, and other supporting materials to: **Dr. Vincent Eckhart, Plant Biology Search Committee, Department of Biology, Grinnell College, 1116 8th Avenue, Grinnell, IA 50112-1690 (telephone: 641-269-4354; fax: 641-269-4285; e-mail: biologysearch@grinnell.edu).** Canddates should also arrange for three letters of recommendation to be sent the mailing address above. To be assured full consideration, all materials should be received by October 16, 2006. For further information about Grinnell College, see our **website: http://www.grinnell.edu.** *Grinnell College is an Equal Opportunity/Affirmative Action Employer committed to attracting and retaining highly qualified individuals who collectively reflect the diversity of the nation. No applicant shall be discriminated against on the basis of race, national or ethnic origin, age, gender, sexual orientation, marital status, religion, creed, or disability.*

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### FACULTY POSITION
**Physiology and Neurobiology**

The Department of Physiology and Neurobiology at the University of Connecticut, Storrs, invites applications for a tenure-track faculty position available in fall 2007, at the **ASSISTANT** or **ASSOCIATE PROFESSOR** level. The successful candidate will be expected to maintain an independent and vigorous research program and participate in the Department's graduate and undergraduate teaching. We encourage applications from individuals studying fundamental physiological or neural processes at the molecular, cellular, or systems level. Applicants must possess a Ph.D. or equivalent and have completed at least two years of postdoctoral training. Candidates for Associate Professor are expected to have a currently funded and active research program. Review of candidates will begin on October 1, 2006, and the search will continue until the position is filled. Send curriculum vitae, a brief summary of current research with a statement of research directions, a statement of teaching interests, and the names of at least three references to: **Chair, PNB Search Committee, University of Connecticut, Department of Physiology and Neurobiology, Box U-3156, 75 North Eagleville Road, Storrs, CT 06269-3156. Website: http://www.pnb.uconn.edu.**

## POSITIONS OPEN

### BIOLOGICAL CHEMISTRY FACULTY POSITION
**Wayne State University**

The Department of Chemistry at Wayne State University seeks applications for a tenure-track position in the Division of Biological Chemistry. Preference will be given toward candidates at the **ASSISTANT PROFESSOR** level. Candidates must have a Ph.D. and the potential to develop a nationally recognized, externally funded research program of outstanding quality in any area of biological chemistry. The Department offers exciting opportunities for candidates with research interests complementing a large group of faculty working in the areas of DNA, RNA, and protein biochemistry, enzymology, carcinogenesis, biophysical, bioorganic, and bioinorganic chemistry, as well as molecular and cellular biology (see Departmental **website: http://chem.wayne.edu** for further information).

The Department of Chemistry has a supportive academic environment and a strong graduate program. Excellent opportunities exist for collaborative research with individuals in the Department of Biological Sciences, the basic science departments in the highly ranked School of Medicine, the College of Pharmacy and Health Sciences, as well as in the Center for Molecular Medicine and Genetics, the Institute for Environmental Health Sciences, and the Barbara Ann Karmanos Cancer Institute. The Department of Chemistry offers an excellent research environment that includes ample, newly renovated research laboratories and a fully staffed Central Instrument Facility that manages state-of the art equipment for: electrospray ionization, and MALDI-TOF mass spectrometry, circular dichroism, electron paramagnetic resonance, surface plasmon resonance, transmission electron microscopy, and nuclear magnetic resonance (NMR), including a 700 megahertz NMR with cryoprobe. The Wayne State faculty also have access to the resources of the Michigan Core Technology Alliance **(website: http://www.ctaalliance.org/),** which includes facilities for bioinformatics, proteomics, genomics, animal models and structural biology, including 900 megahertz NMR and a dedicated synchrotron beamline for X-ray crystallography.

Applicants should submit a complete resume and description of future research plans, as well as three letters of recommendation addressing both research and teaching potential. All materials should be sent to: **Professor Charles H. Winter, Associate Chair, 141 Chemistry, Wayne State University, 5101 Cass Avenue, Detroit, MI 48202-3489.** Review of applications will begin in October 2006. *Women and minority candidates are encouraged to apply. Wayne State University is an Equal Opportunity and Affirmative Action Employer.*

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### FACULTY POSITION
**Department of Ecology and Evolutionary Biology**
**University of Arizona**

A tenure-track position is available as part of a broad initiative in microbial sciences at the University of Arizona. We seek applicants with independent research programs in microbial biology, as exemplified by any of the following areas: (1) microbial evolution and ecology; (2) microbial genomics and/or metagenomic analysis; (3) evolution and/or population biology of infectious disease agents; (4) bioinformatics, computational or evolutionary genomics; (5) systems biology and proteomics in microbial systems. Research organisms can be bacterial, archaeal, or eukaryotic. The start date is flexible but may be as early as summer 2007. Curriculum vitae and statements of research and teaching interests must be submitted online at **website: http://www.uacareertrack.com** (job 35890). In addition, please arrange to have three letters of recommendation sent to: **Amanda Burke, Microbial Biologist Search, Ecology and Evolutionary Biology Department BSW 310, 1041 E. Lowell, University of Arizona, Tucson AZ 85721.** Review of applications will begin October 31, 2006, and continue until position is filled. *The University of Arizona is an Equal Employment Opportunity/Affirmative Action Employer, Minorities/Women/Persons with Disabilities/Veterans.*

## POSITIONS OPEN

### FACULTY POSITIONS
**Smilow Neuroscience Program**
**New York University**
**School of Medicine**

The Smilow Neuroscience Program, a new initiative at New York University (NYU) School of Medicine, seeks to establish an interactive research environment of developmental, molecularn and systems **NEUROSCIENTISTS.** It is housed in the newly dedicated Smilow Research Center, with state-of-the-art facilities. The program is interested in recruiting faculty that combine techniques from multiple disciplines in a novel manner to address fundamental questions of central nervous system circuitry and physiology.

Available positions are tenure track and at the level of **ASSISTANT** or **ASSOCIATE PROFESSOR.** Depending on the area of interest, successful candidates may be affiliated with the Departments of Otolaryngology, Physiology and Neuroscience, or Cell Biology.

Interested individuals are encouraged to visit **website: http://www.med.nyu.edu/xy/yz/html** to learn more about the Smilow Neuroscience Program and the application process. Qualified candidates should apply by following the instructions available on the website.

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### ASSISTANT PROFESSOR IN BIOLOGY

The Chemistry Department of the U. S. Naval Academy invites applications for a tenure-track position in biology at the Assistant Professor level to begin no later than August 2007. The Department consists of 37 full-time faculty members and occupies over 52,000 square feet of newly renovated office, classroom, and laboratory space equipped with a wide array of modern instrumentation and computer facilities. Approximately 30 students per year graduate with American Chemical Society-certified chemistry degrees, and planning is underway for a biochemistry option. The successful applicant must be strongly committed to teaching at the undergraduate level and will be expected to help develop and teach biology and possibly biochemistry courses on a rotating basis with five other biology/biochemistry faculty. The ability to teach general chemistry is also desirable. In addition, the candidate of choice will be expected to develop and maintain a vigorous research program which includes supervision of undergraduate researchers. Candidates should send curriculum vitae, statement of teaching philosophy, concise description of research/scholarly interests, and arrange for three letters of recommendation (at least one of which addresses teaching) to be sent by October 15, 2006, to: **Search Committee, Chemistry Department, U. S. Naval Academy, 572 Holloway Road, Annapolis, MD 21402-5026.** *The U. S. Naval Academy is committed to identifying minority persons and women with the appropriate qualifications and is an Equal Opportunity/Affirmative Action Employer. This agency provides reasonable accommodations to applicants with disabilities.*

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### TWO ASSISTANT PROFESSOR POSITIONS

The University of Wisconsin, Green Bay, invites applications for two tenure-track positions in the Human Biology Department beginning August 2007. First position: **PHYSIOLOGIST.** Teaching responsibilities include human physiology laboratory, anatomy and physiology, and other courses in area of specialty. Second position: **DEVELOPMENTAL BIOLOGIST.** Teaching responsibilities include developmental biology, an introductory biology course, and other courses in area of specialty. Requirements include earned Ph.D. in a biological science and undergraduate teaching experience. Details describing each position may be found at the following **website: http://www.uwgb.edu/hr/jobs.** To insure consideration please submit all application materials by October 15, 2006.

## POSITIONS OPEN

**ASSISTANT PROFESSOR, PURDUE UNIVERSITY, SOYBEAN GENETICS, DEPARTMENT OF AGRONOMY** (tenure-track, academic year appointment), Purdue University, West Lafayette, Indiana (Purdue posting 001657-2006). Position available May 2007.

The Department of Agronomy at Purdue University seeks to fill a tenure-track faculty position in soybean genetics, to further strengthen a comprehensive plant genetics program. The successful candidate is expected to apply basic genetic and genomic tools to translate soybean discoveries to end users. Research foci could include genetics of seed biology, bioenergy, crop enhancement for human nutrition, tolerance to biotic and abiotic stresses, and/or molecular/quantitative approaches that creatively exploit available genetic data for germplasm enhancement. Although previous research experience in the genetics of soybean is desired, individuals with excellent research accomplishments in the genetics or genomics of other plant systems are also strongly encouraged to apply. There are excellent opportunities to collaborate with a strong plant genetics community both locally and internationally. In addition, Departmental searches are underway for a Chaired position in plant genetics and a USDA-Agriculture Research Service adjunct faculty position in soybean genetics/biochemistry. The successful candidate will be expected to teach undergraduate and graduate courses in genetics, mentor students, participate in outreach activities, and be committed to fostering diversity.

Candidates must have earned a Ph.D. in biology, genetics, plant breeding, molecular biology, or related field. Interest in and/or experience in the international dimensions of crop genetics is desirable.

Salary is commensurate with education, training, and professional experience. Excellent fringe benefit package that includes TIAA-CREF retirement program, medical, life and disability insurance, and sabbatical leave program.

Qualified individuals are invited to send a letter of application, including a statement of research goals, teaching philosophy, and curriculum vitae to: **Dr. Craig Beyrouty, Head, Department of Agronomy, Purdue University, 915 West State Street, West Lafayette, IN 47907-2054; telephone: 765-494-4774.** Applications will be reviewed beginning October 15, 2006, and continue until a successful candidate is identified. For more information, please contact: **Search Committee Chair, Scott Jackson (e-mail: sjackson@purdue.edu)** or **Department Head, Craig Beyrouty (e-mail: beyrouty@purdue.edu).** *Purdue University is an Affirmative Action/Equal Access/Equal Opportunity Employer. Women and individuals in underrepresented groups are encouraged to apply.*

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### FACULTY OPENINGS
**Boston College**
**Chemistry Department**

The Department of Chemistry at Boston College invites applications for two Faculty positions to be effective in the fall of 2007: chemical biology and biochemistry and all related interdisciplinary chemistry fields (bioorganic, biophysical, and bioinorganic chemistry); experimental physical chemistry, all branches thereof, and all related interdisciplinary chemistry fields (materials, soft condensed matter).

Applicants are sought at the **ASSISTANT PROFESSOR** level; however, outstanding applicants at the senior level are also welcome. Successful applicants are expected to establish a prominent, externally funded research program, and will join a Department of approximately 125 doctoral students, 25 postdoctoral fellows, 40 undergraduate majors, and an internationally recognized faculty. Boston College, located in a residential community bordering the city of Boston, is within 20 minutes of the major universities and medical centers in the Boston/Cambridge area. For application details, please refer to **website: http://chemserv.bc.edu.** Due date for all applications: 15 October 2006. *Boston College, a university of eight schools and colleges, is an Equal Opportunity Employer and supports Affirmative Action.*

## POSITIONS OPEN

UIC University of Illinois at Chicago

### ASSISTANT OR ASSOCIATE PROFESSOR
### Structural Biology

The Center for Pharmaceutical Biotechnology and the Department of Medicinal Chemistry and Pharmacognosy invite tenure-track faculty applications using structural biology approaches in areas complementary to Center and Department strengths that include both high-profile initiatives in drug discovery and innovative research programs at the interface of chemistry and biology. Ph.D. and postdoctoral experience required. Responsibilities include developing a strong, externally funded research program, and teaching in graduate and professional programs. Candidates at the Associate level must have a strong record of extramural funding. Successful candidate will have joint appointments within the Center and the Department, with extensive collaborative opportunities in a major health sciences center. Forward curriculum vitae, description of research interests, and three reference letters to: **Dr. M. Johnson, Director, Center for Pharmaceutical Biotechnology, College of Pharmacy, University of Illinois at Chicago, 900 S. Ashland, M/C 870, Chicago, IL 60607-7173.** For fullest consideration, please submit all materials by November 1, 2006.

*The University of Illinois is an Affirmative Action/Equal Opportunity Employer.*

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### VERTEBRATE BIOLOGIST
### ASSISTANT PROFESSOR
### Whitman College

**BIOLOGY** (Vertebrate Biologist). Whitman College seeks a full-time tenure-track Assistant Professor of biology, starting August 2007. Ph.D. required; postdoctoral experience preferred. We seek a Vertebrate Biologist with interests in anatomy, evolution, and/or ecology, to teach courses in vertebrate anatomy and general ecology, contribute to an introductory course, The Biological World, and supervise student research and thesis preparation. (Information on the Whitman biology program and course offerings is available at **website: http://www.whitman.edu/biology.)** Whitman College, located in historic Walla Walla near the Blue Mountains in eastern Washington state, has a strong commitment to undergraduate teaching and research in a liberal arts environment. Send (as hardcopy): curriculum vitae, three letters of recommendation, brief statements on teaching and research interests, and transcripts (college and graduate) to: **Vertebrate Biology Search, Biology Department, Whitman College, 345 Boyer Avenue, Walla Walla, WA 99362.** Deadline: October 13, 2006. *Diversity, broadly defined, is a core value of Whitman College; candidates are therefore encouraged to address in their application how they can contribute to enhancing the inclusiveness of the Whitman community.*

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### ENVIRONMENTAL SCIENTIST AND CHAIR

The Environmental Studies Department of Macalester College seeks candidates specializing in environmental science to serve as Department Chair, starting fall 2007. Appointment will be at the **ASSOCIATE** or **FULL PROFESSOR** rank. Areas of expertise could include physical geography, climatology, biogeography, agriculture, or natural resource management, applied mathematics/modeling, environmental chemistry, or other areas. We seek applicants who can provide leadership, help develop and implement a new curriculum, and have experience in an environmental studies/science program. Send letter of application, curriculum vitae, statements of teaching philosophy and research plans, and three letters of reference to: **Dr. Dan Hornbach, Acting Chair, Department of Environmental Studies, Macalester College, St. Paul, MN 55105.** Applications received by November 15, 2006, will receive first consideration. More information is at **website: http://www.macalester.edu/provost/positions/.** *Macalester College is an Equal Opportunity/Affirmative Action Employer and strongly encourages applications from women and minorities.*

## POSITIONS OPEN

**TENURE-TRACK PHYSIOLOGY POSITION.** The Schools of Medicine and Nursing at the University of Missouri, Kansas City, invite applications for three tenure-track positions at the rank of **ASSISTANT** or **ASSOCIATE PROFESSOR.** Two positions will be primary appointments in medicine and one in nursing. The Schools will give preference to candidates with research expertise preferably in cardiovascular and/or exercise physiology to complement existing programs in Women's Health and Cardiovascular Outcomes plus basic research in preeclampsia, cardiovascular imaging, inflammatory cardiovascular disease (plaque/calcification of the coronary arteries and aortic valve) and shock/trauma. Additional opportunities exist for collaboration with basic scientists in the Schools of Medicine, Nursing, Pharmacy, Dentistry and Biological Sciences. Because of our newly established life sciences initiative, additional collaborative research opportunities exist at the nearby Stowers Institute for Medical Research and the Kansas University Schools of Medicine and Nursing. Ample startup funds are available to promote research with high potential for future extramural support. Teaching is expected in the Schools' innovative programs according to the applicant's expertise in nursing, undergraduate or graduate physiology, and in a newly developed Anesthesiology Assistant program. The faculty member selected for these positions will be expected to: (a) teach and mentor students at all levels; (b) maintain an active program of scholarship and funded research; and, (c) provide service appropriate to the position. Required qualifications include: (a) an earned doctorate in physiology, cell biology, nursing,or related field; (b) a record of scholarship, quality teaching and service appropriate to the rank; (c) a history of extramural research support or demonstrated potential to develop a funded research program; and, (d) excellent interpersonal and communication skills. Contact: Applicants should submit a personal letter of interest detailing qualifications, including curriculum vitae, along with names of five references including telephone numbers and e-mail addresses to: **Physiology Search Committee, University of Missouri-Kansas City, School of Medicine, 2411 Holmes Street M1-214C, Kansas City, MO 64108** or **e-mail: gilmoreac@umkc.edu.** Application deadline: Applications will be accepted until the position is filled. *The University of Missouri, Kansas City is an Affirmative Action/Equal Opportunity Employer institution.*

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### FISH ECOLOGY, EVOLUTION, and/or SYSTEMATICS

The Department of Ecology and Evolutionary Biology at the University of Tennessee (UT), Knoxville, seeks to fill a tenure-track position in ichthyology at the **ASSISTANT** or **ASSOCIATE PROFESSOR** level, to start August 1, 2007. Teaching duties will include an undergraduate ichthyology course and a discipline-specific graduate course. A major responsibility of this position is supervision and further development of the superb UT fish collections. Successful applicants will have demonstrated the ability to interact and collaborate broadly in their research and teaching. An earned Ph.D. in a relevant field is required. Postdoctoral or faculty experience is preferred, and applicants will be expected to develop an externally funded and internationally recognized research program. For more information visit **website: http://eeb.bio.utk.edu.** The University welcomes and honors people of all races, genders, creeds, cultures, and sexual orientations, and values intellectual curiosity, pursuit of knowledge, and academic freedom and integrity. Candidates should apply to: **Dr. Edward E. Schilling, Department of Ecology and Evolutionary Biology, 569 Dabney Hall, University of Tennessee, Knoxville, TN 37996.** Applicants should send curriculum vitae, statements of research and teaching goals, and arrange for three reference letters to be submitted. Applications will be reviewed beginning October 20, 2006.

*The University of Tennessee is an Equal Employment Opportunity/Affirmative Action/Title VI/Title IX/Section 504/ADA/ADEA institution in the provision of its education and employment programs and services.*

## POSITIONS OPEN

University of Michigan Medical School

### FACULTY
### Molecular Oncology
### The University Of Michigan Medical School

The Department of Pathology is seeking Tenure-Track Faculty to further build our research program in molecular oncology. Areas of particular interest include but are not limited to epigenetic regulation of transcription, protein modeling/chemical genomics, and genomic and proteomic approaches to biomarker discovery.

The Department, Medical School, and Health Care System are all in excellent financial condition. Over 350 faculty actively participate in the University of Michigan Comprehensive Cancer Center. The Department operates its own graduate program and has nine endowed Chairs and over $20 million annually in research expenditures. Particular areas of research strength include molecular oncology, aging, immunology, proteomics, and informatics. The newly created Divisions of Pathology Informatics and Translational Pathology Planning offer cutting edge technologies in support of research programs. Planning for new building to house the Department's clinical, research, and educational activities is currently underway.

The successful applicant will hold a Ph.D. or M.D./Ph.D. and direct a vigorous research program supported by external funding. Ample resources are available to qualified applicants. Academic rank will be on the tenure-track with rank commensurate with experience. Qualified applicants should submit a letter of interest, summary of research interests, curriculum vitae, and names of three references to:

**Jay L. Hess, M.D., Ph.D.**
**Carl V. Weller Professor and Chair**
**Department of Pathology**
**University of Michigan Medical School**
**Medical Science I Building, Room M5242**
**1301 Catherine Road**
**Ann Arbor, MI 48109-0602**

*The University of Michigan Health System is an Affirmative Action Employer and welcomes applications from women and minorities.*

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### BIOLOGY AND PHYSICS FACULTY POSITIONS

Tenure-track, **ASSISTANT PROFESSOR** positions in evolutionary developmental biology, microbiology, urban ecology, physics and physics education, will be available in the Department of Biological and Physical Sciences at Kennesaw State University beginning August 2007. Applicants should have a strong potential for developing an externally funded research program involving undergraduates. Preference will be given to applicants with demonstrated excellence in teaching at the college level. An earned doctorate in an appropriate discipline is required. For a complete description of positions, go to **website: http://www.kennesaw.edu/facultypositions/.** Review of applications will commence on 13 October 2006, and will continue until the position is filled. Submit a letter describing qualifications for the position, a statement of teaching philosophy, a statement of research interests, current curriculum vitae, graduate transcripts, and the names, addresses, telephone numbers, and e-mail addresses of three references to: **Dr. Dale Vogelien, Evolutionary Developmental Biology Search Committee; Dr. Jerald Hendrix, Microbiology Search Committee; Dr. Heather Sutton, Urban Ecology Search Committee; Dr. Ted LaRosa, Physics Search Committee;** or **Dr. Taha Mzoughi, Physics Education Search Committee, Department of Biological and Physical Sciences, #1202, Kennesaw State University, 1000 Chastain Road, Kennesaw, GA 30144-5591.** *Affirmative Action/Equal Opportunity Employer. Women and minorities are encouraged to apply.*

## POSITIONS OPEN

### ASSISTANT PROFESSORSHIPS IN BIOLOGY
**Western Washington University**

The Biology Department at Western Washington University, a regional comprehensive university located between Seattle and Vancouver, British Columbia, invites applications for two tenure-track, Assistant Professor positions, beginning September 2007. We seek individuals committed to undergraduate and M.S. education who will establish vigorous research programs that involve students. **QUANTITATIVE GENETICIST:** Ph.D. in genetics or evolutionary biology and postdoctoral experience required. Applicants must have training in quantitative genetics and provide evidence of the ability to teach upper-level courses in general genetics and evolutionary biology. Applicants who investigate evolutionary or ecological questions using quantitative genetics are of particular interest. **CELL PHYSIOLOGIST:** Ph.D. and postdoctoral experience in cell physiology required, preferably in an animal system. The applicant must provide evidence of the ability to teach introductory and advanced courses in cell biology and physiology. Applicants who investigate structure-function relationships or integrated regulation of function at the cell, tissue, or organ level are of particular interest. See full position announcements, including all required qualifications, at **website: http://biol.wwu.edu/biology/.** To apply, submit curriculum vitae, statements of teaching and research interests, and three letters of reference. Review begins November 1, 2006. All materials should be sent to the attention of: **Dr. Carol Trent, Chair: Quantitative Geneticist Search Committee; Dr. David Leaf, Chair: Cell Physiologist Search Committee; Biology Department, Western Washington University, 516 High Street, Bellingham, WA 98225-9160.** *Affirmative Action/Equal Opportunity Employer.*

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As part of a continuing expansion program, the School of Chemistry and Biochemistry of the Georgia Institute of Technology seeks to fill several tenure-track faculty positions. Exceptional candidates in particular in the following areas are encouraged to apply: (1) inorganic chemistry; (2) macromolecular structural biology; (3) organic chemistry; and (4) bio-analytical chemistry. Outstanding candidates at all ranks are encouraged to apply; candidates with interdisciplinary interests will be considered for joint appointments with other departments. Further information is available at **website: http://www.chemistry.gatech.edu.** Entry level candidates should send an application letter, curriculum vitae, a summary of research plans, and three letters of reference. Advanced candidates should send curriculum vitae and the names of three references. All materials and requests for information should be directed to: **Chair of the Search Committee, School of Chemistry and Biochemistry, Georgia Institute of Technology, Atlanta, GA 30332-0400.** Applications will be considered beginning October 13, 2006. Applications received past that date will be considered until the positions are filled. *Georgia Tech is an Equal Education/Employment Opportunity Institution.*

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### FACULTY POSITION
**in Biochemical and Biological Engineering**

The Mork Family Department of Chemical Engineering and Materials Science of the University of Southern California (USC) Viterbi School of Engineering is interested in recruiting faculty with expertise in the areas of biochemical and biological engineering. In addition to teaching in the Department's programs, the candidate is expected to develop a strong research program in this area. Significant opportunities exist for collaboration and interdisciplinary research within the Viterbi School, the USC Keck School of Medicine, the USC National Science Foundation Engineering Research Center in Biomimetic MicroElectronic Systems, and the Alfred Mann Institute. Qualified applicants should contact **Professor Theodore Tsotsis** by **telephone: 213-740-2227,** or by **e-mail: tsotsis@usc.edu.** *USC is an Affirmative Action/Equal Opportunity Employer and strongly encourages applications from women and members of underrepresented groups.*

## POSITIONS OPEN

### Biology Faculty Opportunities

Elizabethtown College, located in Central Pennsylvania, currently has two openings in our Biology Department: **ASSISTANT PROFESSOR** of molecular biology and **ASSISTANT PROFESSOR** of physiology. Both positions require a Ph.D. Please visit **website: http://www.etown.edu/humanresources** for full ad and application instructions. *Affirmative Action/Equal Opportunity Employer.*

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### UNIVERSITY OF PITTSBURGH
**Department of Psychology**

The Department of Psychology of the University of Pittsburgh announces a tenure-track position at the **ASSISTANT PROFESSOR** level (pending budgetary approval) in human behavioral genetics/neurogenetics.

Candidates for this position should incorporate molecular and/or quantitative genetic approaches into a biologically-informed research program targeted towards understanding the associations among genes, brain, and behavior. The area of specialization should fall within the scope of one or more of our graduate training areas in biological and health, clinical, cognitive, developmental, or social psychology. For more information about these specialty areas and existing programs of faculty research, see **website: http://www.pitt.edu/~psych.**

The University has a broad range of outstanding resources and established collaborations that facilitate research in genetics and neuroscience, including those involving the Department of Human Genetics **(website: http://www.hgen.pitt.edu),** Center for the Neural Basis of Cognition **(website: http://www.cnbc.cmu.edu),** the Center for Neuroscience at the University of Pittsburgh **(website: http://cnup.neurobio.pitt.edu),** and the Department of Psychiatry **(website: http://www.wpic.pitt.edu).**

The review of applications will begin immediately, with applications received by November 1, 2006, receiving full consideration. Interested parties should submit a cover letter, a research and teaching statement, three letters of recommendation, representative publications, and curriculum vitae to: **Genetics Search, Department of Psychology, University of Pittsburgh, 210 South Bouquet Street, 3129 Sennott Square, Pittsburgh PA, 15260.** For more information about the position, please contact **Anthony Caggiula** at **telephone: 412-624-4501 (e-mail: tonypsy@pitt.edu).**

*The University of Pittsburgh is an Affirmative Action, Equal Opportunity Employer. Women and members of minority groups underrepresented in academia are especially encouraged to apply.*

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### PHYSICAL CHEMISTRY FACULTY POSITION
**University of California, Los Angeles (UCLA)**

The Department of Chemistry and Biochemistry of the University of California, Los Angeles (UCLA), intends to make a tenure-track faculty appointment in physical chemistry (either experimental or theoretical). Candidates at all ranks will be considered. Candidates must give evidence of exceptional promise (for a junior appointment) or great distinction (for a senior appointment) in research and teaching. Applications should include curriculum vitae, a statement of research accomplishments and description of proposed research (not exceeding four pages), reprints of representative publications, and a list of professional references. Junior faculty applicants should arrange to have three letters of recommendation sent at the time of application. To assure consideration, all application materials should be received by October 31, 2006, and directed to:

**Chair**
**Physical Chemistry Search Committee**
**Department of Chemistry and Biochemistry**
**University of California, Los Angeles**
**P.O. Box 951569**
**Los Angeles, CA 90095-1569**
**Fax: (310) 206-8010**

*UCLA is an Equal Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.*

## POSITIONS OPEN

### FACULTY POSITION
**Physics and Astronomy**
**University of Pennsylvania**

The Department of Physics and Astronomy at the University of Pennsylvania invites applications for a junior faculty appointment in the area of experimental biological physics. We are particularly interested in scientists who will pursue research programs that complement and enhance Penn's university-wide nanoscience/nanotechnology initiative. In addition to joining Departmental efforts in cell signaling, single-molecule studies, molecular self-assembly, and biosensors, the successful candidate will enjoy the benefits of numerous interdisciplinary centers including the Laboratory for Research on the Structure of Matter, the Institute for Medicine and Engineering, the Penn Muscle Institute, and the Nano-Bio Interface Center. Applicants should send a resume including a statement of research interests and accomplishments to: **Professor Tom Lubensky, Chair, Department of Physics and Astronomy, University of Pennsylvania, 209 S. 33rd Street, Philadelphia, PA 19104-6396.** Candidates should also arrange to have three letters of recommendation sent to the same address. Applications will begin to be considered as of December 15, 2006, but the Department will accept applications until the position is filled. *The University of Pennsylvania is an Equal Opportunity/Affirmative Action Employer. Applications from underrepresented groups are particularly encouraged.*

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### FACULTY POSITION IN ORGANIC CHEMISTRY
**University of California, Davis**

The Chemistry Department **(website: http://www.chem.ucdavis.edu/)** at the University of California, Davis, invites applications for one faculty position at the **ASSISTANT PROFESSOR** level in organic chemistry. The preferred candidate will develop a strong research program in organic chemistry at the biological interface. A Ph.D. or equivalent in chemistry, medicinal chemistry, or pharmaceutical sciences is required. The candidate must also demonstrate a strong commitment to undergraduate and graduate teaching. This position is open until filled; but to assure full consideration, applications should be received no later than September 22, 2006. The targeted start date is July 1, 2007. Interested candidates should arrange for three letters of recommendation and both paper and electronic (PDF format) copies of curriculum vitae, publication list, teaching statement, and research plans to be sent to:

**Organic Search Committee**
**Department of Chemistry**
**University of California at Davis**
**One Shields Avenue**
**Davis, CA 95616**

*The University of California is an Affirmative Action/Equal Opportunity Employer.*

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### DEPARTMENT OF BIOCHEMISTRY
**Stanford University**
**School of Medicine**

Applications or nominations are invited for an **ASSISTANT PROFESSOR** position in the Department of Biochemistry. Applicants should have an established record of excellence in original research and teaching. The predominant criterion for appointment in the University tenure line is a major commitment to research and teaching. Candidates should submit curriculum vitae including a list of publications, a description of their research interests, and names, addresses, and telephone numbers of three references to the: **Search Committee Chairman, Department of Biochemistry, Stanford University School of Medicine, 279 Campus Drive Room B400, Stanford, CA 94305-5307.** *Stanford University is an Equal Opportunity Employer and is committed to increasing the diversity of its faculty. It welcomes nominations of and applications from women and members of minority groups, as well as others who would bring additional dimensions to the University's research, teaching, and clinical missions.*

## POSITIONS OPEN

### TENURE-TRACK ASSISTANT PROFESSOR
**Molecular Biology**
**Brandeis University**

The Brandeis Biology Department is seeking to fill a tenure-track position in the broad area of eukaryotic gene expression, beginning fall 2007. We are particularly interested in candidates who study mechanistic aspects of transcriptional regulation, with a focus on nucleic acids or chromatin. We are looking to complement existing strengths at Brandeis in post-transcriptional gene regulation, development and function of the nervous system, chromosome structure and function, biophysics, and structural biology. We expect that the appointment will be made at the Assistant Professor level, although a more advanced appointment for candidates with exceptional qualifications may be considered. Candidates should have a Ph.D., M.D., or both as well as postdoctoral experience. First consideration will be given to applications received by October 15, 2006. Candidates can submit initial information online at **website: http://www.bio.brandeis.edu/facultySearch/appFormMB.php.** Applicants should submit curriculum vitae, research plan, and arrange for three letters of recommendation to be submitted preferably by e-mail to **e-mail: volencenter@courier.brandeis.edu** or in hardcopy to:

**Molecular Biology Search Committee**
**Department of Biology, M.S. 008**
**Brandeis University**
**415 South Street**
**Waltham, MA 02454-9110**

*Brandeis University is an Equal Opportunity Employer, committed to building a culturally diverse intellectual community, and strongly encourages applications from women and minorities.*

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### TENURE-TRACK POSITION
**Plant Ecology**

Connecticut College invites applications for a joint plant ecology appointment in botany and environmental studies. The successful candidate will teach plant ecology, systematic botany, introductory environmental studies, participate in a team-taught introductory ecology course and develop a research program that includes undergraduates. Applicants who can integrate geographic information system or mathematical modeling in their classes and research with students are particularly encouraged to apply. The Connecticut College Arboretum includes 450 acres of collections, natural areas, and experimental areas, all used for teaching and research. Connecticut College is a highly selective liberal arts institution committed to interdisciplinary teaching and research. Applications should include a cover letter; curriculum vitae; copies of transcripts; statements about teaching philosophy and research interests; and three letters of reference sent directly to: **Plant Ecology Search Chair, P.O. Box 5362, Connecticut College, 270 Mohegan Avenue, New London, CT 06320.** Review of applications will begin December 1, 2006. See **website: http://www.conncoll.edu** for more information. *The College is an Affirmative Action/Equal Opportunity Employer committed to developing diversity and sustaining a diverse faculty and staff.*

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### PENNSYLVANIA STATE UNIVERSITY

Penn State Beaver, Penn State Delaware County, and Penn State Hazleton invite applications for faculty positions in biology (tenure-track, **ASSISTANT PROFESSOR,** 36 weeks) to begin August 2007. Teach various undergraduate biology courses using traditional and blended delivery modes and oversee laboratory activities. Publish in refereed journals and provide service. Qualifications: Ph.D. in biology. Delaware County prefers Molecular Biologist with some plant experience; Hazleton prefers specialty in environmental biology. To learn more about the campuses and Pennsylvania State, visit **website: http://www.psu.edu/ur/cmpcoll.html.** To learn more about each position and how to apply, visit **website: http://www.ohr.psu.edu/emplment/staff.htm** and follow the faculty link. *Affirmative Action/Equal Opportunity Employer.*

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### STRUCTURAL BIOLOGY FACULTY POSITION

The Structural Biology Program at Florida State University (FSU) invites applications to fill a tenure-track faculty **ASSISTANT PROFESSOR** position beginning July 1, 2007, or later. FSU is entering a period of aggressive growth in the sciences and technology with additional positions in structural biology and related areas expected in the coming years. Applicants should have research interests that complement and extend existing Program strengths in membrane proteins, contractile proteins, protein dynamics, computational structural biology, and protein-nucleic acid interactions. Candidates must have a Ph.D., postdoctoral training, the ability to communicate effectively, and a record indicating outstanding potential for an independent research career. The successful candidate is expected to develop and maintain a vigorous research program supported by extramural funding, to train graduate students, and to participate in undergraduate and graduate level teaching. Laboratory and office space will be in the Kasha Laboratory with departmental affiliation in the Department of Chemistry and Biochemistry. Send curriculum vitae, concise description of planned research, teaching statement, and three letters of recommendation to: **Dr. Timothy Logan, Structural Biology Search Committee, Institute of Molecular Biophysics, Kasha Laboratory, Florida State University, Tallahassee, FL 32306-4380.** Review of applications will begin October 16, 2006, and continue until the position is filled. *Florida State University is an Equal Opportunity/Access/Affirmative Action Employer.*

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### INORGANIC FACULTY POSITION
**University of California, Los Angeles (UCLA)**

The Department of Chemistry and Biochemistry of the University of California, Los Angeles (UCLA), invites applications for a faculty position in inorganic chemistry at the **ASSISTANT, ASSOCIATE,** or **FULL PROFESSOR** rank. We are seeking a candidate who will establish a vigorous and innovative research program. Depending on the level, candidates must give evidence of potential or demonstrated distinction in scholarship and teaching. Applications should include curriculum vitae, a statement of research accomplishments and proposed research plans (not exceeding four pages), and reprints of representative publications. Applicants at the Assistant Professor level should also arrange for three letters of recommendation to be mailed to the address below. To assure consideration, all application materials and letters should be received by October 31, 2006, and directed to:

**Chair**
**Inorganic Search Committee**
**Department of Chemistry and Biochemistry**
**University of California, Los Angeles**
**P.O. Box 951569**
**Los Angeles, CA 90095-1569**
**Fax: 310-206-8010**

*UCLA is an Equal Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.*

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### ASSISTANT/ASSOCIATE PROFESSOR
**Department of Pharmaceutical Sciences**
**The Feik School of Pharmacy**
**University of the Incarnate Word**
**San Antonio, Texas**

A 12-month tenure-track pharmacology **FACULTY** position in the Department of Pharmaceutical Sciences; preferred start date is January 2007. Candidate for this position should have a Ph.D. in pharmacology, teaching and/or postdoctoral experience is preferred. The candidate will be expected to develop and teach the pharmacology section of the integrated pharmacotherapy courses in the Pharm.D. program.

See our website at **website: http://jobs.uiw.edu** for further information and how to apply. *The University of the Incarnate Word is an Equal Opportunity/Affirmative Action Educator and Employer.*

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### TENURE-TRACK FACULTY POSITION
**in Molecular Microbiology**

The Division of Cell Biology and Biophysics, School of Biological Sciences, University of Missouri, Kansas City, invites applications for a full-time tenure-track faculty position at the **ASSISTANT PROFESSOR** or **ASSOCIATE PROFESSOR** level. Preference will be given to individuals with research and teaching expertise in molecular microbiology. The successful candidate will be required to establish a strong research program compatible with one of the School's focus areas of molecular cell biology or structural biology. We seek an outstanding scholar with demonstrable achievements in research, teaching experience in an English-language institution, and exemplary communication and supervisory skills. Teaching will be in at least two of the following subject areas: bacteriology, molecular biology, cell biology, virology, biochemistry. State-of-the-art core facilities are maintained by the School, and competitive salary, startup funds, and laboratory space will be provided. Review of applications will begin immediately and continue until the position is filled. Applications, including curriculum vitae, reprints of publications, summary of present and future research plans and three letters of recommendation (to be solicited by the applicant), should be forwarded to: **CBB Search Committee, Division of Cell Biology and Biophysics - BSB 403, University of Missouri-Kansas City, 5100 Rockhill Road, Kansas City, MO 64110-2499.**

*Equal Opportunity/Affirmative Action Employer*

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### INORGANIC CHEMISTRY
**Brandeis University**

The Department of Chemistry invites applications for a faculty position in inorganic chemistry at the **ASSISTANT PROFESSOR LEVEL.** Outstanding senior candidates will also be considered. Potential areas of interest include, but are not limited to, organometallic chemistry, materials, catalysis, and bioinorganic chemistry. Candidates are expected to establish an innovative research program and display a commitment to undergraduate and graduate teaching. The appointment is planned to commence on or after July 1, 2007. Applicants must submit curriculum vitae and a description of their research plans and arrange for three letters of recommendation (or, for senior applicants, include a list of referees) to be sent to: **Inorganic Search Committee, Department of Chemistry, MS-015, Brandeis University, 415 South Street, Waltham, MA 02454-9110** or **e-mail: chmsrch@brandeis.edu.** For information about the Department, visit **website: http://www.chem.brandeis.edu.** Applications will be reviewed as they become complete, but first consideration will be given to applications received by November 15, 2006. *Brandeis University is an Equal Opportunity Employer, committed to building a culturally diverse intellectual community, and strongly encourages applications from women and minority candidates.*

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### BIOCHEMISTRY FACULTY POST

Bucknell University seeks outstanding candidates for an entry-level, tenure-track position at the rank of **ASSISTANT PROFESSOR** scheduled to begin in August 2007. The successful applicant will teach biochemistry, lecture and laboratory, and will also teach introductory organic chemistry, general chemistry, or introductory inorganic chemistry, depending on background and interests. The successful applicant will be expected to develop a vigorous research program in some area of biochemistry (including bioorganic, bioinorganic, and chemical biology), and this research program will involve undergraduate and Master's students. Ph.D. required, postdoctoral experience preferred. Applicants should send curriculum vitae, summary of research and teaching interests, and three letters of recommendation to: **Professor Timothy G. Strein, Chair of the Biochemistry Search Committee, Department of Chemistry, Bucknell University, Lewisburg, PA, 17837** by October 13, 2006. *Bucknell University encourages applications from women and members of minority groups (Equal Employment Opportunity/Affirmative Action).*

## POSITIONS OPEN

### ASSISTANT/ASSOCIATE/FULL PROFESSOR
**Cancer Biology or Drug Therapy**

The Department of Pharmaceutical Sciences, Texas Tech University Health Sciences Center (TTUHSC), seeks applicants for an attractive new tenure-track faculty position at the Assistant/Associate/Full Professor level. The successful candidate will join a vibrant and expanding group of extramurally funded biomedical and pharmaceutical cancer researchers **(website: http://www.ttuhsc.edu/sop/pharmsci)** who are part of a newly developed university cancer institute. Applicants must have a doctoral degree with research experience in any aspect of cancer biology, chemotherapeutic drug resistance, anticancer drug development or delivery. The successful candidate will be expected to develop an extramurally funded research program as well as teach Pharm.D. and graduate Ph.D. students. Candidates with current NIH funding who have training in pharmacology or drug therapy are particularly encouraged to apply. Competitive salary, startup package, and laboratory space are available. Applicants should submit documents online at **website: http://jobs.texastech.edu** (job requisition 61308). Please include curriculum vitae, a summary of research interests, and names and addresses of three references. TTUHSC in Amarillo includes the School of Pharmacy, School of Medicine, and the Harrington Cancer Research Center. The Department has 23 full-time faculty with interests in cancer biology, brain/vascular, and pharmaceutical research, and state-of-the-art core facilities. For questions, contact the **Search Committee Chair, Dr. U.S. Rao. E-mail: us.rao@ttuhsc.edu. Telephone: 806-356-4015, ext. 294.** *TTUHSC is an Equal Opportunity/Affirmative Action Institution. Minorities and women are encouraged to apply.*

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### WATER/WETLANDS SCIENTIST
**University of Mississippi**

The Center for Water and Wetland Resources seeks an **ASSOCIATE DIRECTOR** and **RESEARCH ASSISTANT/ASSOCIATE PROFESSOR.** The successful candidate will assist the Director in administering the Center, conduct funded research primarily at the University of Mississippi field station, and teach one course and one seminar/year at the field station. The Center has new teaching and research facilities at the 750-acre field station, located 11 miles from campus. The Station has forests, meadows, and 220 experimental ponds of 0.1 to 2 acres.

For information about the Center, field station and complete job description see **website: http://www.baysprings.olemiss.edu.** To apply, send a cover letter outlining research experiences and interests, curriculum vitae, and contact information for three references to: **Dr. Ray Highsmith, Director, Center for Water and Wetland Resources, University of Mississippi Field Station, 15 CR 2078, Abbeville, MS 38601.**

Review of applications will begin on September 18, 2006, and continue until the position is filled. For questions not addressed by this ad or the website, please call **telephone: 662-915-5479.**

*The University of Mississippi is an Equal Opportunity, Affirmative Action Employer.*

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**BIOLOGY ASSISTANT PROFESSOR.** Hamline University invites applications for a tenure-track position to begin September 2007. We seek candidates with research interests in developmental biology or neurobiology. Applicants must be committed to teaching and developing an active undergraduate research program. Postdoctoral experience and ability to contribute to a scientific computing emphasis in the Science Division desired. Ph.D. required. For complete information see **website: http://science.hamline.edu.** Send cover letter, curriculum vitae, teaching philosophy, research plans, and three reference letters to: **Bonnie Ploger, Biology Department, Hamline University, 1536 Hewitt Avenue, St. Paul, MN 55104,** or **e-mail: bploger@hamline.edu.** Application review begins October 9, 2006. *Members of underrepresented groups are strongly encouraged to apply. Hamline University is an Equal Educational/Employment Institution.*

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### FACULTY POSITIONS
**Cell and Molecular Biology and Neurobiology**
**Boston University**

The Biology Department invites applications for two tenure-track appointments at the **ASSISTANT PROFESSOR** level. Exceptional senior applicants will also be considered. New laboratory facilities in an interdisciplinary Life Science and Engineering Building and attractive startup packages are offered. Responsibilities will include establishing an independent research program with extramural funding and active participation in undergraduate and graduate teaching.

Cell and molecular biology: We seek a colleague who conducts research in any area of modern cell and molecular biology, but seek to complement expertise in developmental genetics, cell signaling, gene regulation, molecular motors, protein trafficking, and oncogenesis.

Neurobiology: We seek a colleague who uses a molecular/cellular approach to study basic neurobiological questions in any established animal model system. Research areas include (but would not be limited to): development, sensory mechanisms, plasticity, mRNA or protein localization, or cellular signal transduction in a neural system.

By November 1, 2006, please submit curriculum vitae, a statement of research and teaching interests, representative reprints, and three letters of reference. Submit either electronically to **e-mail: dejames@bu.edu,** or by hardcopy to: **Dr. Ulla Hansen, Chair, Cell and Molecular Biology Search Committee** or to: **Dr. Michael Baum, Chair, Neurobiology Search Committee, Department of Biology, Boston University, 5 Cummington Street, Boston, MA 02215.**

Please visit the following website for information about the Biology Department **(website: http://www.bu.edu/biology/).** *Boston University is an Equal Opportunity/Affirmative Action Employer.*

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### DEVELOPMENTAL BIOLOGY
**Tenure-Track Position**
**Bowdoin College**

The Biology Department at Bowdoin College invites applications for a tenure-track position in developmental biology at the **ASSISTANT PROFESSOR** level beginning fall 2007. We are seeking candidates who will demonstrate excellence in both teaching and research. Postdoctoral experience preferred. Typical teaching responsibilities each year include one laboratory course in developmental biology (with a laboratory instructor), one course at the nonmajors or introductory biology level, and one advanced course in one's area of research. The successful applicant is expected to pursue an active research program that involves undergraduates.

Review of applications will begin November 1, 2006, and will continue until the position is filled. Please send curriculum vitae and a description of your research interests and teaching philosophy, and arrange to have three letters of reference sent to: **Search Committee Chair, Biology Department, 6500 College Station, Bowdoin College, Brunswick, ME 04011-8465.** For further information about the college, the Department, and the program, please see our **website: http://academic.bowdoin.edu/biology/.**

*Bowdoin College is committed to equality through Affirmative Action and is an Equal Opportunity Employer. We encourage inquiries from candidates who will enrich and contribute to the cultural and ethnic diversity of our college. Bowdoin College does not discriminate on the basis of age, race, creed, color, religion, marital status, gender, sexual orientation, veteran status, national origin, or disability status in employment, or in our education programs.*

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**DEAN.** Seattle University invites applications for the position of Dean of the College of Science and Engineering. For further information on the position responsibility and application procedure, please visit **website: http://www.seattleu.edu/scieng/search/ad/** where detailed information is provided. Review will begin on October 15, 2006, and will continue until the position is filled. *Seattle University is an Equal Opportunity, Affirmative Action Employer.*

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### ASSISTANT PROFESSOR
**Evolutionary Biology**

The Department of Biological Sciences seeks candidates with outstanding research that employs modern analytical methods in the study of fundamental aspects of the evolutionary process. Areas of specialization may include field and/or laboratory studies on molecular aspects of population genetics, molecular mechanisms of phenotypic expression, cell division, asexual or sexual development, neural/endocrine processes, genome conservation, or phylogeny. The successful candidate for this **TENURE-TRACK** position will have the potential or demonstrated ability to generate extramural funding and have a commitment to instructional excellence at the undergraduate and graduate levels. The College of Arts and Sciences at Lehigh is especially interested in qualified candidates who can contribute, through their research, teaching, and/or service, to the diversity and excellence of the academic community. Applications should be directed to: **Professor M. Itzkowitz, Chair, Evolutionary Biology Search Committee. E-mail: inbios@lehigh.edu.** Send curriculum vitae, representative publications, description of research and teaching interests, and four letters of reference to the **Search Committee Chair** electronically or to: **Department of Biological Sciences, 111 Research Drive, Lehigh University, Bethlehem, PA 18015.** Deadline for submission is December 1, 2006.

*Lehigh University is an Equal Opportunity Affirmative Action Employer and is committed to recruiting and retaining women and minorities.*

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### MICROBIOLOGIST
**University of Mississippi**

The Seabed Technology Research Center, National Institute for Undersea Science and Technology seeks a Marine Microbiologist to be the **ASSOCIATE DIRECTOR** of the Center and **RESEARCH ASSISTANT/ASSOCIATE PROFESSOR.** The successful candidate will assist the Director in administering the Center, provide leadership in a marine microbial observatory program, and maintain an extramurally funded research program.

A complete job description is at **website: http://www.baysprings.olemiss.edu.** To apply, send a cover letter outlining research experiences and interests, curriculum vitae, and contact information for three references to: **Dr. R. C. Highsmith, National Institute for Undersea Science and Technology Executive Director, Univeristy of Mississippi Field Station, 15 CR 2078, Abbeville, MS 38601.**

Review of applications will begin on September 18, 2006, and continue until the position is filled. For questions not addressed by this ad or the website, please call **telephone: 662-915-5479.**

*The University of Mississippi is an Equal Opportunity, Affirmative Action Employer.*

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### TENURE-TRACK FACULTY POSITIONS
**in Gastroenterology**

The Division of Gastroenterology and Hepatology at the University of Rochester School of Medicine is recruiting faculty at the **ASSISTANT, ASSOCIATE,** and **FULL PROFESSOR** level for tenure-track positions. We are interested in M.D.s and/or Ph.D.s whose primary research is focused on gastrointestinal inflammation. Specific areas of focus might include inflammatory bowel diseases and mucosal immunology, hepatic inflammation and immunology, inflammation in carcinogenesis, vascular inflammation, obesity-related inflammation, and pancreatic inflammation/exocrine function. Generous startup packages and laboratory space are available to qualified candidates.

Please send curriculum vitae, a brief statement of research plans, and names of three references to: **Richard G. Farmer, M.D., Chief, Division of Gastroenterology and Hepatology, P. O. Box 646, University of Rochester School of Medicine, 601 Elmwood Avenue, Rochester, NY 14642.**

*The University of Rochester is an Equal Opportunity Employer.*

## POSITIONS OPEN

### ASSISTANT/ASSOCIATE PROFESSOR
**Division of Pharmacology**
**University of Missouri, Kansas City**
**School of Pharmacy**

The Division of Pharmacology in the School of Pharmacy invites applications for a 12-month, tenured or tenure-track position at the Assistant/Associate Professor level. Applicants should possess a Ph.D., Pharm.D. or M.D. in pharmacology, neuroscience, toxicology, or a related discipline. Preference will be accorded to applicants with interdisciplinary research experience involving neuroscience, pharmacogenomics, substance abuse or translational research; outstanding candidates from other relevant areas are also strongly encouraged to apply. The successful applicant at the Associate Professor level is expected to have a vigorous, well-established, and externally funded research program; and to provide instruction in the Doctor of Pharmacy professional program and dental pharmacology program. The position includes excellent compensation, startup package, and comprehensive benefits. Application review will begin immediately, and will continue until the position is filled.

University of Missouri, Kansas City (UMKC) is a comprehensive research university exemplifying the values of education first, innovation, accountability, diversity, and collaboration. More about UMKC is at **website: http://www.umkc.edu,** or go to **website: http://www.umkc.edu/pharmacy.**

Applicants should electronically submit a cover letter and curriculum vitae with research plan, and arrange to have letters from three professional references e-mailed to:

**Anil Kumar, Ph.D., Chair, Search Committee**
**Division of Pharmacology**
**University of Missouri-Kansas City**
**2411 Holmes Street**
**Kansas City, MO 64110-2741**
**Telephone: 816-235-2415**
**E-mail: kumaran@umkc.edu**

*UMKC is an Affirmative Action/Equal Opportunity Institution.*

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Texas Christian University (TCU) invites nominations/applications for its prestigious **ROBERT A. WELCH CHAIR IN CHEMISTRY.** While the area of research is open, we seek a scholar with an internationally recognized research program that will complement existing faculty strengths. Previous Welch Chairs at TCU include **Paul D. Bartlett** and **C. David Gutsche.**

Located in the Fort Worth/ Dallas area, TCU is an independent, coeducational institution of approximately 7,200 undergraduate students and 1,500 graduate students, offering 98 undergraduate majors and 20 graduate degrees in 59 areas, including six doctoral fields of study, with a commitment to both teaching and research. The Chemistry Department offers the Ph.D., M.S., and B.S. degrees and is well equipped, particularly with excellent X-ray diffraction, mass spectroscopy, and nuclear magnetic resonance facilities. Please send curriculum vitae, detailed research plans, and a list of references to: **Professor Jeffery L. Coffer, Department of Chemistry, P.O. Box 298860, Texas Christian University, Fort Worth, TX 76129.** E-mail inquiries may be sent to **e-mail: j. coffer@tcu.edu.** The search is ongoing until the position is filled. *TCU is an Equal Employment Opportunity/Affirmative Action Employer.*

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### ASSISTANT PROFESSOR
**University of California, Santa Cruz**

The Earth and Planetary Sciences Department seeks applicants for a **TENURE-TRACK** position in geobiology. Position available: July 1, 2007. Open until filled. For full consideration, applications must be received by November 22, 2006. Refer to position 686-07. For full details, see our **website: http://www.es.ucsc.edu/jobs/index.html** or contact **Judy Van Leuven, e-mail: judy@pmc.ucsc.edu; telephone: 831-459-4478.** *Affirmative Action/Equal Employment Opportunity Employer.*

## POSITIONS OPEN

### POSITION ANNOUNCEMENT

Molecular Studies of Virology and/or Immunology, the Department of Veterinary Pathobiology, Center for Veterinary Health Sciences (CVHS), Oklahoma State University (OSU) invites applications for a tenure-track research faculty position in infectious diseases at the rank of **ASSISTANT** or **ASSOCIATE PROFESSOR.** Applications are encouraged from individuals with training/interests in virology and/or immunology with emphasis in viral immunity. Candidates with interests and training in RNA viruses are encouraged to apply. Applicants must have a Ph.D. degree and relevant postdoctoral experience. Persons with the D.V.M. degree as well are encouraged to apply. Responsibilities include the development of a strong extramurally funded research program utilizing modern, molecular approaches to solving problems relating to animal or human diseases, and participation in the CVHS graduate education program. The CVHS has a strong commitment to a well-funded program in comparative medicine, and this position should provide leadership and support for that program. Opportunities exist for collaboration with other research faculty in the CVHS, Oklahoma Agricultural Experiment Station, University of Oklahoma Health Sciences Center, Oklahoma Medical Research Foundation, in addition to other OSU departments involved with molecular genetics and infectious disease research. Interested individuals should send an application including curriculum vitae, statement of professional goals, and names of three references to: **Dr. Robert W. Fulton, Search Committee Chair, Department of Veterinary Pathobiology, CVHS, Stillwater, OK 74078, telephone: 405-744-8170.** To ensure full consideration, applications should be received by December 15, 2006, and review of applications will continue until a suitable candidate is identified.

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### CHAIR, BIOCHEMISTRY AND CELL BIOLOGY
**Rice University**

The Biochemistry and Cell Biology Department at Rice University **(website:http://biochem.rice.edu/)** invites applications for **DEPARTMENT CHAIR.** We are seeking a dynamic scientist with an outstanding research program within the broad area of biochemistry and cell biology and a strong interest in graduate and undergraduate education to lead in the implementation of Rice University's vision for the second century **(website: http://www.professor.rice.edu/professor/10_Points.asp).** The ideal candidate will bring innovative ideas, enthusiasm, and acumen for success, and will engage the Department toward a shared goal of outstanding international research recognition. Applicants should submit curriculum vitae, brief statement of research interests, and the names of three references to: **George Bennett, Chair, Search Committee, Biochemistry and Cell Biology, MS-140, Rice University, P.O. Box 1892, Houston TX 77251-1892. E-mail: gbennett@rice.edu.** *Rice University is an Equal Opportunity/Affirmative Action Employer. Women and minority candidates are particularly encouraged to apply.*

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### TWO TENURE-TRACK VIROLOGY POSITIONS

The Biomedical Sciences and Pathobiology Department of the Virginia-Maryland Regional College of Veterinary Medicine **(website: http://www.vetmed.vt.edu/Organization/Departments/DBSP/index.asp)** invites applications for two tenure-track faculty positions (rank open) in the area of viral pathogenesis and replication as part of an interdisciplinary program focused on emerging and re-emerging infectious diseases. See further information and submit applications via the Virginia Tech **website: http://https://jobs.vt.edu,** using posting number 183918. Fill out the application form and upload: cover letter, curriculum vitae, contact information for three references, and statement of research goals. Review begins August 1, 2006, but positions are open until filled. *Virginia Tech is an Equal Opportunity/Affirmative Action Employer.*

## POSITIONS OPEN

### CHAIR, DEPARTMENT OF BIOCHEMISTRY AND MOLECULAR BIOLOGY
**University of South Alabama**
**College of Medicine**

The University of South Alabama College of Medicine is seeking a Chair for the Department of Biochemistry and Molecular Biology. The successful candidate will be an outstanding, nationally recognized scientist and academician who will recruit new faculty and set the future direction for the Department's research and educational missions. Candidates with a strong record of research in any area related to biochemistry and/or molecular biology will be considered. Excellent interpersonal skills, leadership, and commitment to mentoring junior faculty and trainees are essential. Credentials appropriate for the rank of tenured Professor are required. Current research focus in the Department includes molecular and cellular signaling; more information is available at **website: http://southmed.usouthal.edu/com/biochem/.** Opportunities for collaboration and program development exist within the Institution, including the Center for Lung Biology, the Mitchell Cancer Institute, and other departments in the College of Medicine. The Department contributes to medical education and to the training of graduate students through the interdisciplinary Ph.D. program in Basic Medical Sciences. The University is located in Mobile on Alabama's Gulf Coast.

Interested applicants should submit curriculum vitae, names and contact information for at least three references, a statement of research interests and academic vision, and a summary of administrative experience either electronically **(e-mail: mtownsley@usouthal.edu)** or by mail: **Dr. Mary Townsley, Chair, Biochemistry Chair Search Committee, Dean's office, CSAB 170, University of South Alabama, College of Medicine, Mobile, Alabama 36688.** Review of applications will begin October 16, 2006, and continue until the position is filled.

*The University of South Alabama is an Affirmative Action/Equal Opportunity Employer.*

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**BIOLOGY.** Linfield College seeks applicants for a tenure-track **ASSISTANT PROFESSOR** with a specialization in community or ecosystem level ecology beginning July 1, 2007. Four courses taught annually include: an ecology course with a laboratory or field component for biology and environmental studies majors; an additional course with laboratory or field component for biology majors; participation in an introductory course for biology majors; a nonmajors course in area of specialty. Successful applicants will demonstrate a commitment to, and potential for, developing a vigorous research program with undergraduates. Ph.D. in biology or related field required; postdoctoral experience preferred. Send application letter, statements of teaching philosophy and research interests specific to this position, official transcripts of all college and university work, teaching evaluations (if available), three letters of reference, and curriculum vitae by October 31, 2006, to: **Dr. J. Christopher Gaiser, Linfield College, Unit A468, 900 S.E. Baker, McMinnville, OR 97128.** Additional information regarding this position may be found at **website: https://www.linfield.edu/humanresources/teaching.php.** *Equal Opportunity Employer.*

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**BIOCHEMISTRY FACULTY POSITION.** University of Wisconsin, Oshkosh, seeks tenure-track **ASSISTANT PROFESSOR** beginning September 1, 2007. Requirements: Ph.D. in biochemistry. Responsibilities: teach undergraduate biochemistry, general chemistry, and laboratories in organic chemistry; advise chemistry majors; establish an active research program in biochemistry; and pursue extramural funding. Submit: letter of application, curriculum vitae, three current confidential letters of recommendation, transcripts, research plans, and one-page statement of teaching philosophy to: **Dr. C.P. Gibson, Chair, Chemistry Department, University of Wisconsin Oshkosh, Oshkosh, WI 54901.** (E-mail applications not accepted.) Application deadline: October 27, 2006. *Affirmative Action/Equal Opportunity Employer.*

## POSITIONS OPEN

### FACULTY POSITION(S)
**Department of Chemistry and Biomolecular Science**
**Clarkson University**

Outstanding, energetic candidates are sought to fill two tenure-track faculty positions in the Department of Chemistry and Biomolecular Science. The search is aimed at entry-level **ASSISTANT PROFESSOR** candidates, but exceptionally qualified applicants may be considered for more senior appointments. Research expertise in all areas of the broadly defined biomolecular and biomaterials sciences will be considered. Teaching will be in support of the Department's rapidly developing program in biomolecular science, and could include such courses as biochemistry, medicinal chemistry, biomaterials, et cetera. Depending on specialization, the position could be closely associated with the University's New York State-supported Center for Advanced Materials Processing. Candidates must have a Ph.D. with outstanding research potential and be capable of teaching courses at the Ph.D., M.S., and undergraduate levels. Successful candidates are expected to develop vigorous, creative, externally funded research programs. Please submit vitae, research and teaching plans, and list of references to: **Sergiy Minko, Chair, Search Committee, Department of Chemistry and Biomolecular Science, Clarkson University, Potsdam, NY 13699-5810.** Positions start fall 2007. Review starts immediately and will continue until filled. Position posting 20-06 and 23-06. *Clarkson is an Affirmative Action/Equal Opportunity Employer.*

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**BIOCHEMISTRY.** Baylor University announces a faculty position, **ASSISTANT PROFESSOR** of biochemistry, beginning fall 2007. The Department is housed in a new $103 million science facility, part of an exciting plan of growth at the University. Any area of biochemistry will be considered. The successful candidate will enjoy many opportunities for teaching and research in a dynamic environment that fosters interdisciplinary collaboration. Requirements: Ph.D. in biochemistry, chemistry, or closely related field; commitment to exemplary teaching at both the undergraduate and graduate level; and potential for a vigorous, independent, externally funded research program. Postdoctoral experience is desirable. Send letter of application, full curriculum vitae, summary of future research plans, estimate of startup costs, undergraduate and graduate transcripts, and three letters of reference to: **Chair, Search Committee, Department of Chemistry and Biochemistry, One Bear Place #97348, Baylor University, Waco, TX 76798-7348. E-mail: mary_lynn_trawick@baylor.edu.** Applications will be reviewed beginning September 4, 2006, and will be accepted until the position is filled. To ensure full consideration, your application must be completed by October 25, 2006. Baylor is a Baptist university affiliated with the Baptist General Convention of Texas. *As an Affirmative Action/Equal Employment Opportunity Employer, Baylor encourages minorities, women, veterans, and persons with disabilities to apply.*

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### ASSISTANT PROFESSOR
**Watershed Science / Forest Hydrology**

The University of Missouri is seeking a tenure-track (12-month) faculty member for a teaching and research position. She/he will teach undergraduate courses in watershed management and summer field studies and a graduate course in their specialty. Development of a nationally recognized, extramurally funded research program in a relevant area is expected. Ph.D. required, with one degree in forestry or a comparable area; postdoctoral experience preferred. Send letter of application, resume, transcripts, description of research interests, teaching philosophy, and contact information for three references by October 31, 2006, to: **Dr. R.M. Muzika, Department of Forestry, 203 ABNR Building, University of Missouri, Columbia, MO 65211. Telephone: 573-882-8835, e-mail: muzika@missouri.edu.** Additional information at **website: http://www.snr.missouri.edu/jobs/watershed.html.** *University of Missouri is an Equal Opportunity, Affirmative Action Employer.*

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### FACULTY POSITION IN CELL BIOLOGY
**University of Texas Southwestern Medical Center at Dallas**

The Department of Cell Biology at the University of Texas Southwestern Medical Center at Dallas is seeking to appoint exceptional scientists in the field of cell biology and cellular imaging to the position of **ASSISTANT PROFESSOR** (tenure track). The candidates must have a Ph.D. or M.D. and specialize in research areas at the junction between cell and molecular biology such as; molecular interactions in living cells, cellular basis of tissue organization, and spatial organization of signal transduction. The excellence of the individual candidate will take precedence over the area of special interest. The successful candidate will join an internationally recognized cell biology faculty at a top-rated medical institution and receive both a competitive salary and an exceptional startup package. For more information, visit the cell biology **website: http://www8.utsouthwestern.edu/utsw/cda/dept25128/files/34664.html.** Applicants should e-mail their curriculum vitae, the names of three references, and a brief description of their research goals to the attention of **Dr. Richard G. W. Anderson** at **e-mail: cb.recruitment@utsouthwestern.edu.**

*The University of Texas Southwestern Medical Center is an Affirmative Action/Equal Opportunity Employer. Women and minority candidates are encouraged to apply.*

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### ASSISTANT PROFESSOR
### FERMENTATION BIOLOGIST

The Department of Animal Sciences at the Ohio State University is accepting applications for a 12-month, tenure-track position as a Fermentation Biologist. The incumbent will be expected to compete for extramural funding and strengthen the campus-based teaching curriculum. The Department's website is **website: http://ansci.osu.edu/.** An excellent laboratory and bioreactors are located at the Wooster campus. Candidates will hold a Ph.D. in the life sciences with an emphasis on microbial population dynamics, metabolomics, and (or) chemical processes regulating biomass conversion of agricultural wastes in anaerobic systems. Postdoctoral experience is desired. Applicants should send a statement of interest in the position, curriculum vitae, and the names, postal and e-mail addresses, and telephone numbers of at least three references who may be contacted to: **Dr. Jeff Firkins, Search Committee Chair, Department of Animal Sciences, 2029 Fyffe Court, Columbus, OH 43210. Telephone: 614-688-3089; e-mail: firkins.1@osu.edu.**

*Ohio State University is an Equal Opportunity, Affirmative Action Employer. Women, minorities, Vietnam-era veterans, disabled veterans, and individuals with disabilities are encouraged to apply. Requires successful completion of a criminal background check.*

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### TENURE-TRACK FACULTY POSITION
**Department of Neurobiology and Anatomy**
**Wake Forest University**
**School of Medicine**

The Department of Neurobiology and Anatomy invites applications for a faculty position to join a dynamic and expanding collaborative research group investigating how the brain integrates information from different senses (multisensory integration). Individuals with interests and skills that would complement and/or expand the current investigations are strongly encouraged to respond. The position will be available July 1, 2007. Candidates should send curriculum vitae, a statement of specific research interest, and three letters of recommendation to: **Search Committee for Multisensory Integration Position, Department of Neurobiology and Anatomy, Wake Forest University School of Medicine, Winston-Salem, NC 27157-1010.** For more information on the Department and areas of research emphasis, visit our website at **website: http://www.wfubmc.edu/nba.** *Wake Forest University School of Medicine is an Affirmative Action/Equal Opportunity Employer.*

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### BIOCHEMIST/MOLECULAR BIOLOGIST
**Bryn Mawr College**

The Department of Biology invites applications for a tenure-track faculty position in biochemistry/molecular biology at the rank of **ASSISTANT PROFESSOR.** We are searching for an individual who will thrive in an environment that combines teaching and research. The successful candidate is expected to teach at all levels of the curriculum and establish an externally funded research program that provides rigorous collaborative research projects for undergraduates. Candidates with expertise in genomics and other emerging areas of informatics are encouraged to apply. A doctorate and at least one year of postdoctoral research experience required. Submit curriculum vitae, description of research plans that addresses the role of undergraduates in your research, statement of teaching philosophy, and arrange for three letters of recommendation to be sent by September 15, 2006, to: **Chair, Biology Search, Department of Biology, Bryn Mawr College, 101 N. Merion Avenue, Bryn Mawr, PA 19010-2899.** (No electronic submissions please.)

Located in suburban Philadelphia, Bryn Mawr College is a highly selective liberal arts college for women who share an intense intellectual commitment, a self-directed and purposeful vision of their lives, and a desire to make meaningful contributions to the world. Bryn Mawr comprises an undergraduate college with 1,200 students, as well as coeducational graduate schools in some humanities, sciences, and social work. The College supports faculty excellence in both teaching and research, and participates in consortial programs with the University of Pennsylvania, and Haverford and Swarthmore Colleges. *Bryn Mawr College is an Equal-Opportunity, Affirmative Action Employer. Minority candidates and women are especially encouraged to apply.*

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**TEXAS TECH UNIVERSITY (TTU).** The Department of Chemistry and Biochemistry invites applications for two tenure-track faculty positions at the **ASSISTANT, ASSOCIATE,** or **PROFESSOR** rank. We anticipate hiring in the broadly defined areas of materials chemistry or medicinal chemistry. A Ph.D. is required; postdoctoral experience is preferred. The successful candidate is expected to have or develop an independent, well-funded research program, and have a commitment to excellence in teaching at the undergraduate and graduate levels. Online faculty applications must include curriculum vitae, statement of proposed research (including startup requirements), statement of teaching philosophy, and names of three individuals who have been asked to provide letters of recommendation. Letters of recommendation should be mailed to: **Faculty Search Committee, Texas Tech University, Department of Chemistry and Biochemistry, P.O. Box 41061, Lubbock TX 79409-1061.** Evaluation of applications will begin on October 15, 2006, and continue until the position is filled. Online Faculty Registration for requisition numbers 61766 (materials chemistry) and 61767 (medicinal chemistry) can be found at **website: http://jobs.texastech.edu.** *TTU is an Equal Opportunity/Affirmative Action Institution and actively seeks diversity among its employees.*

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### ASSISTANT PROFESSOR
**The University of Chicago**
**Department of Chemistry**

The Department of Chemistry of the University of Chicago invites applications from outstanding individuals for the position of Assistant Professor of chemistry. This search is in the areas broadly defined as inorganic, organic, and physical chemistry. Applicants must mail hardcopies of curriculum vitae, a list of publications, and a succinct outline of their research plans, and arrange for three letters of recommendation to be sent by mail to: **Michael D. Hopkins, Chairman, Department of Chemistry, The University of Chicago, 5735 S. Ellis Avenue, Chicago, IL 60637.** Review of completed applications will begin October 1, 2006; to ensure full consideration, all material should be submitted by that date.

*An Equal Opportunity/Affirmative Action Employer.*

## POSITIONS OPEN

### ASSISTANT PROFESSOR OF MICROBIOLOGY

The Oklahoma State University (OSU)Center for Veterinary Health Sciences (CVHS) is expanding its research programs in the area of comparative medicine and infectious diseases. A tenure-track position at the Assistant Professor level is available in the area of bacterial pathogenesis/immunology. Preference will be given to individuals whose research focuses on pathogenic bacteria of human and/or animal importance and whose research programs utilize animal models to examine molecular mechanisms of pathogenesis or immune control of bacterial infections. Responsibilities include development of a strong, independent research program supported with extramural funding and participation in the graduate program. Substantial startup support will be available for the successful candidate. Applicants must have a Ph.D. degree and two years of productive postdoctoral research training. While not required, persons also holding a D.V.M. degree are encouraged to apply. OSU CVHS is located in Stillwater, a small city centered around the University. Stillwater provides the opportunity for high quality rural and urban living, a low cost of living, and excellent public schools. Interested persons should send an application including their curriculum vitae, a description of their research interests and career goals, and names of three to five references to: **Dr. R. Eberle, Microbiology Search Committee Chair, Department of Veterinary Pathobiology, Room 250 McElroy Hall, Center for Veterinary Health Sciences, Oklahoma State University, Stillwater, OK 74078.** To ensure full consideration, applications should be received by 15 December 2006. *OSU is an Equal Opportunity Employer.*

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**COMPUTATIONAL BIOCHEMIST, SAN FRANCISCO STATE UNIVERSITY.** The Department of Chemistry and Biochemistry invites applications for a tenure-track faculty position at the **ASSISTANT PROFESSOR** level in computational biochemistry. A successful candidate's education and training will be in chemistry, biochemistry, or related disciplines. Candidates must demonstrate a strong dedication to teaching and research as well as the potential to develop a vigorous, externally funded research program involving undergraduate and M.S. students. Research interests could include but are not limited to molecular dynamics, protein modeling, and protein folding, or computational strategies for studying the proteome. We anticipate that the successful candidate's research will be primarily computational and will provide opportunities to establish collaborations with experimental scientists at San Francisco State University and other academic institutions and industries in the Bay Area. Applicants should send curriculum vitae, a summary of research plans, a statement of teaching philosophy and interests, and three letters of recommendation electronically in PDF to **e-mail: cmpbioch@sfsu.edu,** or by mail to: **Computational Biochemistry Search Committee, Department of Chemistry and Biochemistry, San Francisco State University, 1600 Holloway Avenue, San Francisco, CA 94132.** Further details are available at **website: http://www.chemistry.sfsu.edu.** Review of applications will begin on October 1, 2006. *The University is an Equal Opportunity Employer with a commitment to diversity and encourages applications from women, members of all ethnic groups, veterans, and people with disabilities.*

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### BIOLOGY

**ASSISTANT PROFESSOR,** wildlife biology, tenure-track position at Saint Vincent College beginning August 2007. Teaching responsibilities include general biology, organismal wildlife biology, nonscience majors courses, and supervision of senior research projects. Twelve-credit semester load. Ph.D. and potential for teaching excellence and ongoing research required. See **website: http://www.stvincent.edu/hr2** for full description and application information. Saint Vincent College is a Catholic, Benedictine, liberal arts and sciences college of 1,650 students near Pittsburgh, Pennsylvania.

### MOLECULAR BIOLOGY/MARINE BIOTECHNOLOGY
**University of Mississippi**

The Ocean Biotechnology Center and Repository (OBCR), National Institute for Undersea Science and Technology seeks an **ASSOCIATE CENTER DIRECTOR** and **RESEARCH ASSISTANT/ASSOCIATE PROFESSOR** of pharmacognosy. The research programs of OBCR include marine biotechnology and drug discovery, and environmental health and toxicology. The successful candidate will assist the Director in administering the Center and Repository and will conduct extramurally funded research in marine biotechnology, preferably using molecular techniques.

A complete job description is at **website: http://www.baysprings.olemiss.edu.** To apply, send a cover letter outlining research experiences and interests, curriculum vitae, and contact information for three references to: **Dr. R. C. Highsmith, National Institute for Undersea Science and Technology, Executive Director, University of Mississippi Field Station, 15 CR 2078, Abbeville, MS 38601.**

Review of applications will begin on September 18, 2006, and continue until the position is filled. For questions not addressed by this ad or the website, please call **telephone: 662-915-5479.**

*The University of Mississippi is an Equal Opportunity, Affirmative Action Employer.*

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### FACULTY POSITION
**Biochemistry-Neuroscience**

The Department of Biochemistry at the Weill Medical College of Cornell University invites applications for a full-time, tenured/tenure-track faculty position for individuals with research interests at the interface of neuroscience and biochemistry. All ranks **(ASSISTANT, ASSOCIATE** and **FULL PROFESSOR)** will be considered. The research focus of suitable candidates is broadly defined and includes structural, chemical, biochemical, cell biological, and molecular cellular physiological approaches to fundamental problems of relevance to neuroscience and neurological disease. Interested applicants should forward electronic versions, as Microsoft Word or PDF files, of (1) a covering letter of application; (2) curriculum vitae; (3) an overview of current and future research interests; and (4) the names and contact information of at least three references references to: **The Chair, Search Committee, Biochemistry/Neuroscience, Weill Medical College of Cornell,** to **e-mail: biochem-neuro-search@med.cornell.edu.** Evaluation of applications will commence on October 15, 2006, and applications will be considered until the position is filled. *Equal Opportunity Employer/Minorities/Females/Persons with Disabilities/ Veterans.*

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### FACULTY POSITION IN GENETICS
**Department of Biological Sciences**
**Carnegie Mellon University**

The Department of Biological Sciences at Carnegie Mellon seeks to fill a tenure-track position in genetics. Special consideration will be given to outstanding candidates who study neurogenetics of model organisms. Research areas of current faculty include neuroscience, genetics/molecular biology, cell/developmental biology, computational biology, and biochemistry/biophysics. Carnegie Mellon has a long history of interdisciplinary research with strengths in computational biology, biological imaging, proteomics, and robotics. Candidates must have a doctoral degree and strong research credentials. They will be expected to develop a strong and innovative research program and to participate in the undergraduate and graduate educational programs.

Please send curriculum vitae, statements of research and teaching interests, and three letters of recommendation to: **Dr. A. Javier Lopez, Department of Biological Sciences, Carnegie Mellon University, 4400 Fifth Avenue, Pittsburgh, PA 15213-2683.** Review of applications will begin October 16, 2006. **Website: http://www.cmu.edu/bio.** *Carnegie Mellon is an Equal Opportunity/Affirmative Action Employer.*

### TENURE-TRACK POSITION
**Mammalian Neurobiology or Development**
**Brandeis University**

Brandeis University has an opening for a tenure-track appointment in the Department of Biology and the Center for Behavioral Genomics beginning fall 2007. We seek individuals studying mammalian neurobiology or development using transgenic approaches in mice. A focus on developmental neurobiology is preferred, but mouse geneticists studying any aspect of neuroscience or developmental biology are encouraged to apply. Candidates should have a doctorate as well as postdoctoral experience. An appointment will be made at the level of **ASSISTANT PROFESSOR,** although a more senior appointment is feasible for an outstanding candidate with appropriate experience. First consideration will be given to applications received by November 13, 2006.

Candidates can submit initial information online at **website: http://www.bio.brandeis.edu/facultySearch/appForm.php.** Applicants should submit curriculum vitae and research plan, and arrange for three letters of recommendation to be submitted preferably by e-mail to **e-mail: volencenter@courier.brandeis.edu** or in hardcopy to:

**Developmental Biology Search Committee**
**Department of Biology, MS008**
**Brandeis University**
**415 South Street**
**Waltham, MA 02454-9110**

*Brandeis University is an Equal Opportunity Employer, committed to building a culturally diverse intellectual community and strongly encourages applications from women and minorities.*

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The Department of Biomedical Sciences at the University of South Alabama (USA) seeks candidates for a tenure-track faculty position at the **ASSISTANT** or **ASSOCIATE PROFESSOR** level. The USA Biomedical Sciences Department offers a B.S. degree and is responsible for the undergraduate education of students interested in pursuing postbaccalaureate study in medicine, health professions, or basic sciences. Candidates must have a Ph.D., or equivalent, in one of the biomedical sciences, two years of postdoctoral experience, and expertise to teach human anatomy. The successful candidate will also be expected to develop an active research program and mentor undergraduate research projects. Review of applications will begin on October 15, 2006, and will continue until the position is filled, with an estimated start date of June 1, 2007. Applications should include a cover letter of interest, curriculum vitae, and three letters of reference. The application material should be sent via regular mail or e-mail to: **Dr. Michael P. Spector, Professor, Department of Biomedical Sciences, UCOM 6000, University of South Alabama, Mobile, AL 36688-0002** or **e-mail: mspector@usouthal.edu. Website: http://www.southalabama.edu/alliedhealth/biomedical.** *Affirmative Action/Equal Employment Opportunity/Minorities/Females/Persons with Disabilities.*

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The Department of Applied Health Science at Wheaton College (Illinois) is searching for a **DEPARTMENT CHAIRPERSON** at the **ASSOCIATE PROFESSOR** or **PROFESSOR** level. This is a full-time, tenure-track appointment beginning August of 2007. Applicants must have expertise in the areas of exercise physiology/biochemistry, and/or cardiovascular physiology. The Department is focused on human health and lifestyle.

Applicants should send curriculum vitae and a description of teaching philosophy and research interests to: **Dr. David Ianuzzo, Chair; Applied Health Science; Wheaton College; 501 College Avenue; Wheaton, IL 60187.** Additional application materials will be sent to eligible candidates.

*Wheaton College is an evangelical protestant Christian liberal arts college whose faculty members affirm a Statement of Faith and the moral and lifestyle expectations of our Community Covenant. The College complies with federal and state guidelines of nondiscrimination in employment; women and minorities are encouraged to apply.*

A major initiative in Haemopoietic Stem Cell Biology

## Non-Clinical Chair at King's College London (Denmark Hill Campus)

Department of Haematological Medicine at King's College London is a centre of research and clinical excellence, with one of the largest haematopoietic stem cell transplantation programmes in Europe. In addition to the other School- and College- wide services, there are in-house state of the art facilities, including laser dissection microscopy, transcription profiling, proteomics and GMP laboratories for the production of cell and gene therapy investigational medicinal products for clinical evaluation. The department has an active clinical, translational and basic research programme with a number of specific areas of focus including cell cycle regulation & epigenetics, functional genomics, gene therapy and leukaemia immunobiology.

The holder of this newly established Chair in Haematological Stem Cell Biology will be a research scientist of international standing with an established track record of productivity and research excellence. You will have already made significant contributions to the understanding of the subject as evidenced by seminal publications in the field and peer reviewed grant funding of their research activities. You will be expected to develop and lead an active research programme in one or more aspects of haematopoietic stem cell biology, with a clear emphasis on both fundamental and translational research, including the potential trans-differentiation of haematological stem cells into non-haematopoietic cell lineages. The ultimate objectives being better understanding of the biology and the translation of this knowledge into more effective strategies for the treatment of haematological and other disorders. You will also contribute to the teaching of undergraduates and postgraduate students.

This position is supported by an attractive start-up package including laboratory space for up to 12 investigators within the Rayne Institute at the Denmark Hill Campus.

**To receive an application pack** please e-mail medicine2@kcl.ac.uk or write to the Miss Iman Dhedhi, Personnel Department, 4th Floor Capital House, 42 Weston Street, London SE1 3QD. **Alternatively visit** http://www.kcl.ac.uk/jobs where a full job description and person specification are available when you view this advert on the vacancy bulletin. Informal inquiries can be made to Professor Ghulam J Mufti, Head of Department, by e-mail (ghulam.mufti@kcl.ac.uk) or by telephone 020 7346 3080 or to Professor Farzin Farzaneh, by e-mail (farzin.farzaneh@kcl.ac.uk) or by telephone 020 7848 5902. Please quote the appropriate reference number on all correspondence.
**Job reference**: A5/MCD/647/06
**Closing date for applications**: 12 October 2006

KING'S College LONDON
University of London

*Equality of opportunity is College policy*

## SCIENTIFIC STAFF POSITIONS

The **Center for Functional Nano-materials** (CFN) at Brookhaven National Laboratory (BNL) (http://www.bnl.gov/cfn) is pleased to announce new opportunities for scientific and technical employment beginning in the fall of 2007.

In selected cases, openings will be available in January 2007. The CFN is a science-based user facility funded by the US DOE Office of Basic Energy Sciences to provide state-of-the-art capabilities for the fabrication and study of nano-scale materials. The CFN will feature strong in-house scientific programs while offering broad access to its capabilities and collaboration through an active user program. The CFN seeks to impact the nation's energy security through world-class interdisciplinary research in three focus areas, including **Nano-catalysis** (Synthesis and characterization of in situ reactivity of nano-structured catalysts), **Electronic Nano-materials** (Fundamental excitations in low-D materials, especially heterogeneous nanostructures and strongly correlated systems), and **Biological and Soft Nano-materials** (Self-organization in soft and hybrid functional nano-systems). Corresponding state-of-the-art laboratory facilities are being developed in **Nano-patterning, Advanced Electron Microscopy, Proximal Probes, Theory and Computation, Ultra-fast Spectroscopy, Materials Synthesis, and Small Angle Scattering at the National Synchrotron Light Source**.

Full operations are to begin in April 2008. Outstanding individuals are sought in all of the laboratory facility and theme areas at levels ranging from group leader and senior scientist to junior scientist and post doc. A brief summary of selected job areas is given below. A complete list of openings and full details concerning applying for these positions may be found at **http://www.bnl.gov/cfn/jobs**.

**Electro-catalysis/catalysis of in situ systems, including fuel cells, using surface science, electrochemistry, and synchrotron techniques**

**In situ electron microscopy of chemical reactions, oxide nano-particles, and other nano-structured objects**

**Spectroscopic STM of surface reactions/catalysis and nanostructured electronic nanomaterials**

**Real-time LEEM/PEEM of surface processes**

**Inorganic/materials chemistry, nano-particle synthesis**

**Synchrotron-based, soft-matter/biomaterials nanoscience**

**Ultra-fast laser spectroscopy**

**Nano-scale theory and computation**

**Organic/macromolecular chemistry of surfaces, supramolecular assemblies and hybrid nano-systems**

**Single-molecule optical methods for soft-matter/biomaterials**

**Experimental soft matter physics and biophysics**

**Plasma/vacuum processing technology and nano-fabrication**

**Focused ion beam microscopy and related analytical methods**

**Clean room engineer**

Qualifications for these positions include a Ph.D. in the physical or life sciences, a strong record of technical achievement and the demonstrated ability to conduct creative, independent research. Successful candidates will also have excellent communication and interpersonal skills, as well as the ability to work in a team and interact effectively with a broad range of colleagues.

Interested candidates should respond to **http://www.bnl.gov/cfn/jobs**, select the scientific areas of interest, and respond by including a detailed CV, publications list, and the names of three references.

BNL is an equal opportunity employer committed to building and maintaining a diverse workforce. BNL is managed by Brookhaven Science Associates for the U.S. Department of Energy.

BROOKHAVEN NATIONAL LABORATORY
*a passion for discovery.*

**www.bnl.gov**

## FACULTY POSITIONS

### FACULTY POSITION

The Department of Molecular Physiology and Biophysics at Baylor College of Medicine is seeking to recruit a new faculty member at the **ASSISTANT/ASSOCIATE PROFESSOR** level. We are seeking candidates with a strong record of research contributions, the potential to establish an exciting, independent research program, and a commitment to excellence in graduate and medical student education. Research interests of the applicant must include a physiological or biophysical approach to studying cell or organismal biology, and a focus on advanced technologies or the study of disease models is preferred. The Department has a strong commitment to both basic and translational biomedical research and has state-of-the-art facilities for confocal microscopy, mouse MRI, multiphoton imaging, and a core for the creation and analysis of new mouse models. Baylor College of Medicine is a world-renowned research institution with strengths in many areas including mouse genetics, cardiovascular sciences, and neuroscience. Ample opportunities exist for scientific interaction and collaborations within the Department, throughout the College, and within the Texas Medical Center. Located in the heart of the Texas Medical Center, Baylor College of Medicine is in close proximity to the University of Texas Medical School at Houston, the M.D. Anderson Cancer Center, Rice University, and the University of Houston.

Please e-mail your curriculum vitae, the names and contact information for three references, and a description of both your research program and career goals to **e-mail: wehrens@bcm.edu, Attn: Dr. Xander Wehrens, Chair of the Recruitment Committee, Department of Molecular Physiology and Biophysics, Baylor College of Medicine, One Baylor Plaza, BCM335 Houston, TX 77030.**

The deadline for receipt of applications is October 30, 2006.

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### FACULTY POSITIONS, TENURE TRACK
**Department of Psychiatry**

The Department of Psychiatry and the Center for the Study of Traumatic Stress at the Uniformed Services University of the Health Sciences (USUHS) seeks to fill tenure-track neuroscience laboratory research and teaching positions **(ASSISTANT/ASSOCIATE PROFESSOR).** The Department, twenty full-time faculty, seeks to expand ongoing neuroscience research, animal and human, in: stress; anxiety (particularly acute stress responses, posttraumatic stress disorder and dissociation); depression; behavior and drug use. Individuals who hold Ph D. or M.D. degrees and have active fundable research are invited to apply. Send curriculum vitae, description of current and anticipated research, and three references to: **Robert Ursano, M.D., Chairman, Department of Psychiatry, USUHS, 4301 Jones Bridge Road, Bethesda, MD 20814-4799 (e-mail: rursano@usuhs.mil).** Review of applications is ongoing. *The University is an Affirmative Action/Equal Opportunity Employer.*

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**NEUROSCIENTIST.** The Department of Biomedical Sciences at Colorado State University (**website: http://www.cvmbs.colostate.edu/bms/**) seeks a Neuroscientist, preferably with background in systems or integrative neuroscience, to be hired at the **ASSISTANT / ASSOCIATE PROFESSOR** level. Candidates should have a vigorous, extramurally fundable research program and will be expected to teach in undergraduate, graduate, or professional veterinary programs. Applicants must have a Ph.D., D.V.M., M.D., or equivalent. A letter of application, curriculum vitae, and names of three references should be sent electronically or by post to: **Dr. Sue C. Kinnamon, Department of Biomedical Sciences, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, Fort Collins, CO 80523 (e-mail: sue.kinnamon@colostate.edu).** Review of applications will begin October 23, 2006.

*Colorado State University is an Equal Employment Opportunity /Affirmative Action Employer.*

## FACULTY POSITIONS

Bryant University, College of Arts and Sciences, seeks to fill the following tenure-track position beginning August 1, 2007.

**ASSISTANT PROFESSOR, SCIENCE AND TECHNOLOGY.** Candidates should be committed to upholding excellence in teaching, establishing a scholarly publication record, and serving as an active member of the Bryant community. We are seeking a broadly-trained Biologist with a Ph.D. in biology or related field. The Science and Technology Department is a growing Department, with current minors in biotechnology and environmental science. The successful candidate will need to participate in the creation of new degree programs and maintain an active research program involving undergraduates. This position involves teaching biology and chemistry courses and the coordination and expansion of the biotechnology minor.

Forward a letter of application, current curriculum vitae, and a list of three references to the: **Human Resources Office-PROFJO (541), Bryant University, 1150 Douglas Pike, Smithfield, RI 02917.** Materials may be submitted electronically to **e-mail: humanresources@bryant.edu** (Word or PDF attachments).

Bryant University is a selective four-year, residential university with a strong tradition in business education and developing strengths in the liberal arts. We offer a select number of B.A. programs, as well as undergraduate and graduate degrees in business and information technology. We are located fifteen minutes northwest of Providence, one hour from Boston, and three hours from New York City. The University serves approximately 3,000 undergraduates and 500 graduate (Master's-level) students.

*Bryant University is an Equal Employment Opportunity/Affirmative Action Employer, and an institution committed to diversifying its faculty and student body.*

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### TWO FACULTY POSITIONS
**Department of Cell Biology and Neuroscience**
**Montana State University, Bozeman**
**Website: http://www.montana.edu/cbn**

The Department of Cell Biology and Neuroscience at Montana State University invites applications for two tenure-track faculty positions. Candidates at the **ASSISTANT, ASSOCIATE,** or **FULL PROFESSOR** level with research interests that complement Departmental strengths in developmental neuroscience, biophysics, and systems/computational neuroscience will be considered. Both positions will be expected to contribute to teaching at the undergraduate/graduate level and one of the faculty positions will be required to teach medical gross anatomy in the WWAMI (Washington, Wyoming, Alaska, Montana, Idaho) Medical Education Program **(website: http://www.montana.edu/wwwami/).** The candidate(s) must hold a Ph.D. and/or M.D. degree or equivalent. Complete position descriptions and application information can be found at **website: http://www.montana.edu/msuinfo/jobs/faculty/.**

Review of applications will begin October 13, 2006, and will continue until both positions are filled. *ADA/Affirmative Action/Veterans Preference.*

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Find out about jobs before you get your issue. Sign up for customized e-mail notification of jobs at **website: http://www.sciencecareers.org** by clicking on Job Alerts. You can also post your resume (open or confidentially) and check how many employers have viewed your resume at your own convenience.

## POSITIONS OPEN

### POSTDOCTORAL RESEARCH ASSOCIATE

The Medical Department at Brookhaven National Laboratory (BNL) currently has a Postdoctoral Research Associate position available in the Behavioral NeuroPharmacology and Neuroimaging Laboratory. This position will provide research training in rodent neuroimaging models applied to alcoholism, drug abuse, obesity, or attention deficit hyperactivity disorder under an NIH grant. Requirements include a Ph.D. degree in either neuroscience, biopsychology, pharmacology, or biomedical sciences, an interest in the above research themes, behavior and neuroimaging, significant experience in at least one of the following: rodent brain autoradiography or immunohistochemistry, rodent microdialysis, rodent surgery and microsurgery or rodent behavior - operant conditioning self-administration. BNL's relevant major equipment includes: microPET (positron emission tomography), beta imager (autoradiography), 9-Tesla microMRI, microCT, surgical suite, sophisticated behavior monitoring equipment, and molecular biology facilities. BNL is affiliated with State University of New York at Stony Brook. Interested candidates should forward their curriculum vitae and research interests to **e-mail: thanos@bnl.gov** referring to position number M3954 or mail to: **P. Thanos, Brookhaven National Laboratory, Building 490, Upton, NY 11973-5000.** *BNL is an Equal Opportunity Employer committed to workforce diversity.*

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### NEUROGENETICS RESEARCH ASSOCIATE

Position available at Yale University for M.D. or Ph.D. trained in human genetics to join multidisciplinary research program studying genetic channelopathies, with emphasis on pain and other sensory disorders (see **Waxman and Dib-Hajj,** *Trends in Molecular Medicine* **11:**555, 2005; **Han et al.,** *Annal of Neurology* **59:**553, 2006). Superb opportunity to collaborate with Molecular and Cell Biologists, Electrophysiologists, et cetera. Send curriculum vitae, statement of interest, and three letters of reference to: **Stephen G. Waxman, M.D., Ph.D., Chairman, Department of Neurology LCI-707, Yale School of Medicine, P.O. Box 208018, New Haven, CT 06520-8018.** *Yale is an Affirmative Action/Equal Opportunity Employer. Women and minorities are encouraged to apply.*

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### POSTDOCTORAL FELLOWS
**Harvard University**

The six Environmental Fellows at Harvard University who will start work in September 2007, will be outstanding scholars with a doctorate in any field and a research interest in the environment. Each fellow will work with a host faculty member in the host's laboratory or office. Excellent salary and benefits. Apply by January 15, 2007. Details at **website: http://www.environment.harvard.edu.**

*Harvard is an Equal-Opportunity, Affirmative-Action Employer.*

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**STAFF SCIENTIST.** Research glucose transport to determine mechanism by which HIV protease inhibitors induce metabolic syndrome, investigating function of Glut4 glucose transporter. Requires Ph.D. in biology, medical science, related field with focus in glucose transporter molecules; and knowledge of glucose metabolism, Glut1& 4 function/properties, mammalian cell culturing, apoptosis pathways and assays, rDNA. Curriculum vitae to: **Dr. M. Mueckler, Department of Cell Biology and Physiology, Washington University Medical School, 660 S. Euclid, P. O. Box 8228, St. Louis, MO 63110.**